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ALBERT HOLMES SMITH: A STUDY IN COURAGE*

Presidential Address

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THE dilemma of choosing a subject for the time-honored address has evidently been a recurring ailment of some duration. In support of this, I find that when the Society held its meeting in Philadelphia in 1899, President Joseph Tabor Johnson, of Washington, remarked as follows: "The seeker after a suggestion for a theme for a future Presidential address will stand appalled as he glances even at the backs of twenty-three volumes—knowing full well that all the live themes and burning questions have been thoroughly, scientifically, and eloquently discussed." If these were the feelings of an essayist then, what must they be today? Being historically minded, I reviewed the past addresses of this nature, noting that on only two occasions had individual Fellows been chosen as subjects of an address, if my memory is correct. Both were men of unquestionable stature—Marion Sims and Howard Kelly.

In response to this inclination I planned early this year to share with you the story of a Founder and early President of this Society, the richness of whose life was never marked by stellar accomplishments or startling discovery, but whose courage, steadfastness, gentleness, and self-discipline were part and parcel of his character. I first learned of this staunch but modest personage 20 years ago, when I was writing a brochure commemorating the seventieth anniversary of the Obstetrical Society of Philadelphia, and on the basis of this knowledge I have entitled my address, "Albert Holmes Smith: A Study in Courage."

The subject of our address was of distinguished lineage. His forebears were of English extraction; they emigrated from Yorkshire to William Penn's Colony about 1685, as did so many Quakers, and others, too, who suffered

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privation, to worship as they chose. His father, Moses B. Smith, an especially well-qualified physician, was a native of Solebury, Bucks County, Pennsylvania, as was his mother, nee Rachel Coate, also of the Friends' persuasion. He practiced first in the suburban town of Bustleton, but in 1828 moved to the city where he became an intimate of Philip Syng Physick, James, and others of this later Colonial period. Finally, in 1832, he relinquished practice entirely because of an illness that eventually proved fatal—an autopsy revealed a malignant tumor of the right kidney, a fact worthy of remembrance in view of his son's subsequent history. In their home at Sixth and Vine Streets, not far from the majestic Delaware, where the shad ran abundantly in the spring of those days, a third son, the youngest of 7 children, was born on July 19, 1835, and named Albert Holmes Smith.

Nestled among the rolling hills of pleasant countryside in Chester County, where the Brandywine flows nearby, the Westtown Academy came into being as a Friends' boarding school in 1799. There the young lad was sent for a brief period before continuing his education at Gregory's Classical School in Philadelphia. This prepared him well for an A.B. degree awarded him at the University of Pennsylvania, where he delivered the Latin oration at the Commencement in 1853. That fall he entered the Medical School and graduated in 1856 at the age of 21. During his course, he had in the same vein received practical tutelage from the intellectually gifted George Bacon Wood. The latter, a versatile author and historian, was a chemist and pharmacologist, and learned in *materia medica*—incidentally, an art that is now undergoing an invigorating renaissance. Wood held various professorships in several institutions and was President of the College of Physicians of Philadelphia during a period of 31 years—an inspiring teacher for so able and receptive a student as Albert Holmes Smith.

The young graduate sought and secured an appointment immediately as physician to the Frankford Hospital for the Relief of Persons Deprived of Their Reason. While his superior, Dr. Joshua Worthington, was absent on a mission in Europe, young Smith proved a reliable administrator. After 18 months' service, however, he resigned to become a resident physician in the Pennsylvania Hospital, the cornerstone of which had been laid on May 28, 1755, with Benjamin Franklin's inscription reading "In the year of Christ MDCCLV, George the Second, happily reigning." In this respected hospital, Smith came in contact with the outstanding practitioners of that day. James Tyson, as noted in his memoir, testifies that he was "among the best residents the hospital knew," and furthermore he was ever loyal to it.

Smith left the Pennsylvania, when his term ended, with the realization that "taking an office," as one said in those days, was not the final rung in the educational ladder. So, with characteristic determination, he took advantage of the lectures of the talented obstetrician, Joseph Warrington. Twenty years earlier Warrington had organized the Philadelphia Nurse Society to command better nursing and also the "Philadelphia Lying-in Charity Hospital for Attending Women in Their Own Homes," as it had been chartered in 1832. Located originally in older areas of Philadelphia, this venerable institution is well known today as the Lying-In Department of the Pennsylvania Hospital, constructed in 1929. It has been supervised by a succession of Fellows of this Society. The present incumbent (R. A. K.) is with us today.

Retired in 1857 from active work, Warrington was an ideal person to mold the obstetric thinking of his graduate students. Smith was one of these. Seriously impressed with the specialty, he was soon given an assistantship, and was destined, together with his older bachelor friend, Joshua G. Allen, to be made an Attending Physician and Lecturer himself in 1863, continuing in this capacity for 20 years, or until 1882, when illness began to assail him, and Allen

finished his lectures for him. Regarding this long stint of teaching, the comment was made by an observer that "the value of knowledge that he [Smith] imparted to hundreds of young men who are now engaged in obstetric practice all over the Union can scarcely be overestimated." I suspect that some of you, upon hearing this quote, are thinking long, long thoughts in this regard.

The marriage of Albert Holmes Smith and Emily Kaighn, of Kaighn's Point, New Jersey, took place in 1860. He had "married out of meeting," as expressed in that day. The records of the Arch Street Meeting reveal that in 1861 the Meeting was informed of this, which was contrary to their Order of Discipline. When questioned, Smith acknowledged the correctness of the charge, presenting a statement to the committee expressing his "sincere and heartfelt regret that circumstances not under his control prevented his marriage according to our discipline and that we, convinced of the truth of our doctrine and testimony, trust that the Meeting will pass by the violation." Evidently the error in discipline was excused, for an entry in the record some time later states, "A. H. S. has been informed of the conclusion of the Meeting in his case."

The marriage prospered. Of 7 children born, 5 were to survive him, together with his wife. The eldest son was Edward B. whose descendants carry on the family name with credit. The younger son, Major Lawrence Savery Smith (named for his father's staunch friend and loyal supporter, Dr. William Savery) died, while on his way home from the Spanish American War, in 1898, of typhoid fever contracted in Puerto Rico during his service as Regimental Surgeon, First Pennsylvania Volunteer Infantry. Three daughters lived to an advanced age.

During the Civil War years Philadelphia was a large and important center for the care of the sick and wounded. Including local hospital and army installations, 22 such institutions are recorded. Naturally, the Lying-in Charity Hospital was not among them, but one may assume that Albert Smith was kept reasonably busy there during the war years. In any event, during the tragic days of reconstruction we find him established in family life, launched in practice, adequately trained in obstetrics, with a zeal for graduate teaching, and with an exceptional hospital appointment. To this extent fortune had smiled upon him.

In 1867, at the age of 32, Albert Smith took a step that became a symbol of the courage that was to exemplify his career for all time. He accepted the post of Consultant to the Woman's Hospital. In that day and age of the so-called emancipation of women, this was treason indeed, and the wrath of most of his colleagues descended upon him; women doctors were especially despised and a bit of background may well be in order.

Sixteen years before, in 1850, the Female Medical College of Pennsylvania had been founded in a small house in the old Colonial area of Third and Arch Streets and not too far from Franklin's grave in Old Christ Church Yard. A decade later the title was changed to "The Woman's Medical College," and it was soon obvious to its supporters that a woman's hospital must be the next step to complement the medical school's bedside teaching. Fundamentally this was necessary for development, but there was another angle too. Whenever women students ventured into the amphitheater of the Pennsylvania Hospital, and even elsewhere, for purposes of observation, they were taunted and scoffed at by the student body and addressed with sarcasm by the instructor.

Humiliation of this sort could not continue, and on April 11, 1861, a group of vitally interested women met in a home near the college to plan such a hospital, and 3 months later the needed institution was functioning in a converted building in a growing area on the western rim of the city. Dr. Catharine Macfarlane, of Philadelphia, told me that when she was an interne in the

Woman's Hospital in 1898, she "was told that Dr. Smith would sit in the parlor on the first floor while Dr. Croasdale and Dr. Broomall operated upstairs—'just in case.'"

Following these formative years, Albert Smith consistently became a creative figure in Philadelphia's medical world. He worked in a persuasive way and his leadership commanded respect. The qualities of mind, character, and disposition that he exhibited may have stemmed from family tradition and his upbringing in the discipline of the Society of Friends. His courage in adhering to the principles of equal rights for women was manifested constantly by outspoken support of an unpopular cause.

"The reaction of his position regarding women," one narrator remarks, "varied from those who were not prepared to accept his opinions in full, from others partly so but friendly, and from some who actually turned their backs upon him, seeking in every way to defeat his ardent wish in State and County Societies." He was one of a minority that recognized a similar concern, but Albert Holmes Smith, tall, robust of figure, and darkly handsome in countenance, with black hair and hazel eyes, was the one who would meet the onslaught with calm and dignity. His bearing was described as courteous and in every sense that of a gentleman.

I had the good fortune, as did some of my colleagues, to know Daniel Longacre, who died in 1949 in the ninetieth year of his life, having been an active, and later an honorary, member of the Obstetrical Society of Philadelphia for 65 years. He told us many tales of those figures of the past. Of Albert Holmes Smith he said to me years ago, "a nobler man than he never breathed, a greater obstetrician never taught." This is a direct quotation.

Significant of more trials to come was a resolution referred to the Council of the College of Physicians of Philadelphia in the spring of 1868, and definitely directed at Smith. Lengthy and ponderous in tone, the gist of this resolution, uttered by one Dr. Littell, was that "women as a general rule being physically, morally, and intellectually" unable to function as doctors, such association with them must be forbidden, and "that the Fellows are hereby instructed and conjoined to govern themselves in accordance with this declaration, holding no connections with such institutions (i.e., hospitals and colleges) and no professional intercourse with such practitioners." Going through the legislative channels of this time-honored College, Smith moved the adoption of a motion that the Council's opinion be sustained—"that it is not desirable that the college should take any action on the question." This was seconded, passed, and sustained despite the opponent's insistence on an "aye and nay" vote. Nonetheless tradition was so ingrained that 64 years passed before a woman physician became a Fellow. On the evening of Jan. 11, 1932, the late President Francis R. Packard, erudite medical historian, greeted a newly elected Fellow with the words, "You are making history tonight, Dr. Macfarlane."

A dominant characteristic of Albert Smith's was his ability to act definitively when convinced of the worth of an idea to which he had given mature consideration. In this regard, one of four prominent channels of his destined activity came into being when he conceived and organized the Obstetrical Society of Philadelphia in the summer of 1868. In response to his invitation, a number of interested colleagues met at his residence on South Broad Street on the evening of June 6 to discuss the idea. The response was favorable, the initial group was rapidly enlarged, and 2 weeks later the Obstetrical Society of Philadelphia took its rank with the Boston and New York Societies. Naturally, he was most attentive to the Society, contributing to the programs in the usual way and presenting ingenious devices for use in the procedure of those days. The Smith pessary may be regarded as a modification of the Hodge, and

there was a vaginal speculum he designed, but apart from the pessary that bears his name and still maintains its usefulness, other inventions and modifications remain only in the archives. He served 2 terms as president (1874 and 1875), a mark of deserved respect, in spite of differences of opinion.

Most important was the meeting of Nov. 6, 1879, a story told in entirety in a previous publication, and representative of Smith's continued efforts to promote educational advantages for women physicians. Two years before, on Dec. 6, 1877, a paper written by Dr. Emeline Cleveland, a woman of exemplary character, and professor of Obstetrics and Diseases of Women and Children at the Woman's Medical College, was presented for her by Dr. Smith. It related the result of a fistula operation. On that occasion Professor William Goodell, of the University of Pennsylvania, a former President, had opposed the idea, but the publication committee had passed the paper. It was with this precedent in mind that Dr. Smith now made a motion that when a member in good standing announced in advance that he wished to present a paper or specimen of a nonmember physician, that the said nonmember be considered the guest of the Society at that time and be invited by the presiding officer to enter into the discussion. The motion was properly seconded, and then the opposition really let loose. One after another "viewed with alarm." A stellar remark was that this was a steppingstone from women visitors to women members, and that Dr. Smith under the by-laws had no right to introduce them, "as the pronoun 'he' is used a number of times in them, and until a 'she' can become a 'he' the women were excluded by the by-laws as they stand."

Smith answered his opponents calmly and without rancor, arguing that the membership should consider the stand taken upon this question by the medical world and should realize that men everywhere were changing their views on this subject in favor of women's emancipation. But a vote of 18 to 9 defeated his efforts, and not until 1892 were women physicians finally accepted as eligible for election. Nonetheless he continued to encourage the faculty and staff of the Women's Medical College to submit reports of their activities, and he in turn would present them for the essayists, the publication committee would accept them, and the material would then appear in the "Transactions." This was Smith's ingenious approach toward securing recognition of the women's continuing accomplishments.

As the year 1880 approached, disturbed health, as well as added responsibilities, were met with accustomed fortitude, but for some months Smith was out of touch with the Obstetrical Society. He resumed attendance sporadically through 1881-1883; but his last recorded appearance was in May of the latter year, when he reported a case of mumps complicated by peritonitis in a young girl.

Smith's relationship with the Obstetrical Society therefore was understandably close, both with respect to his professional contributions and through its service as a forum for his persistent advocacy of equal opportunities for women.

An equally important facet of responsibility and further testing of his positive views came about during the period of his career in the Philadelphia County Medical Society. He became a member of this Society relatively late in his professional life, for he was 41 when elected in January, 1876. Nonetheless, 4 years later he was installed as President after a strongly contested election in which the same antagonism that faced him first in the College of Physicians and again in the Obstetrical Society came into play again. Unfortunately he had scarcely occupied the chair when he came down with typhoid fever, intermittently epidemic in those days. Pollution of the water supply because of defective drainage and sewage disposal was a constant menace. There was a common jest abroad in that era that citizens were contaminated by the drainage of the cemeteries of their ancestors situated on the hillsides of the Schuylkill River. Report after

report presented by the Medical Society's Committee on Hygiene, and resolutions to the City Council fell on deaf ears as far as positive legislation was concerned. Smith's invalidism continued for 3 months or more and a sea voyage was advised as a health measure. A resolution of the censors wishing him "Bon Voyage" and hoping for his return to health and for his resumption of chairmanship was cheering to him and gratefully acknowledged.

He sailed home in September after having been received with acclaim by his colleagues abroad and entertained by the Gynecological Society of Great Britain. The year 1881 saw him re-elected promptly, an opponent having withdrawn in his favor. His chairmanship was characterized by dignity and courtesy, while his skill as a parliamentarian was of the highest order. During this time, and in practically every meeting, the question of female membership was discussed pro and con. As in the meetings of the Obstetrical Society, the fundamentalists of the opposition presented no argument other than that the by-laws said "he" and that was that! Throughout the entire year, however, Smith, seconded by a group of like-minded supporters, presented propositions of membership regularly for physicians of the Woman's Medical College Faculty. The replies from the censors were ever in kind—"held under advisement" and "not eligible . . . under the by-laws." It was also recorded that it was not even valid to "pass a resolution which shall supplement or change existing by-laws so long as there exists a legal way by which such change can be made."

With respect to organized medicine at the state level, he cooperated with Hiram Corson of Plymouth Meeting in the presentation of resolutions urging the appointment of women to attend female patients in state institutions for the insane on a parity with male physicians. In every instance they were voted down, but by ever smaller majorities. When attending such sessions in Lancaster, Smith drove there daily to support his heartfelt belief. That he returned to Philadelphia later in the day to attend to his patients is indicative of his physical stamina. The biographer, James Tyson, also his personal physician, tells how Smith could lose sleep night after night but work the next day without complaint. Smith confessed that he preferred deliveries at night; it gave him more freedom for the care of his daytime patients.

The fourth sphere of action in Albert Smith's engaging life is rightly concerned with our own Society. It is a dramatic, saddening story, and, in a career as broad as his, interrelated incidents happened that need telling.

His vitality seemed never-ending as he journeyed to New England for the meeting of the American Gynecological Society held in the quarters of the Boston Society of Natural History in September, 1882. Drysdale, of Philadelphia, spoke on his belief in the diagnostic importance of the "ovarian corpuscle" when confronted with ascites or huge abdominal cysts. Samuel Busey, of Georgetown, decried the baneful effects of the high-heeled French shoes upon the health and well-being of the female and her pelvic organs. Smith returned home as one of the 2 newly elected Vice-Presidents. The other was the brilliant and versatile Theophilus Parvin, soon to leave Louisville to take the Chair of Obstetrics and Diseases of Women and Children at Jefferson Medical College. Subsequently, as colder weather came and the days grew shorter, minor discomfort suggestive of prostatic trouble annoyed Albert Smith. Undeterred, he kept his own counsel.

Throughout the new year of 1883, in addition to his professional duties, he continued in regular attendance at the County Society meetings. With perseverance he advanced the proposal of women members—and met criticism with equanimity. A direct descendant of the renowned Atlee brothers of Lancaster declared at one heated session that if women were to be balloted for, he wished to withdraw his son's name from the posted list for election! Came September, and with it the meeting of the American Gynecological Society in Philadelphia, held

in the College of Physicians. Paul Mundé of New York suggested that consideration be given to holding the annual meetings at "various watering places"; evidently Philadelphia weather was not at its best. This session also marked the elevation of Albert Holmes Smith to the presidency. Was it the awareness of his new responsibility that prompted him now to seek medical consultation?

The term "lithotripsy" is defined as "the operation of crushing calculi in the urinary bladder," and Sir Henry Thompson, Professor of Surgery and Pathology in the Royal College of Surgeons in London, was renowned for his skill in performing this operation. Years earlier he had become Surgeon Extraordinary, and had been knighted as well, for his success in operating upon the royal bladder of Leopold I, King of the Belgians. Sir Henry was less successful with Napoleon III, exiled in England after Sedan. Long a sufferer from calculi, the deposed emperor succumbed suddenly a few days after the operation with the Empress Eugenie beside him in their refuge at Chiselhurst. To this famed consultant Albert Smith now turned.

Sir Henry diagnosed adenoma of the prostate gland. He did not think it malignant but there was a possibility that it might become so. Smith returned to America somewhat encouraged, and resumed duties in the new year. Not until April, however, did he put in an appearance at the County Society, and then to protest against an amendment to the by-laws whereby the word "male" was to be inserted in order to make the matter of membership unequivocal once and forever. The proposed amendment failed for want of the necessary two-thirds majority, and a repeat performance met the same fate 2 months later, and for the same reason but with an increased vote against it. This must have encouraged Smith. Could it be that his patient and persuasive attitude was gaining adherents for the cause to which he was now giving his last measure of devotion? Soon thereafter he left for Beach Haven, on the New Jersey coast. There he would rest and prepare for the Gynecological Society meeting in Chicago, where he must journey late in September.

Beach Haven may be known to some of you—a rather small community on narrow Long Beach, flanking inland water between it and the Jersey mainland, with the broad Atlantic at its doorstep. Smith hoped to recuperate at the family cottage there and give thought and substance to the address ahead. He did not feel well, and an attack of malaria had assailed him, but he worked with fortitude nonetheless. Back in Philadelphia, he visited the County Medical Society on September 24, taking with him Dr. Edward Jackson of West Chester. There is no record extant of Smith's further attendance there. A few days later he entrained for Chicago and called the meeting to order at 9:00 A.M., Sept. 30, 1884. William H. Byford of Chicago, a founder and Past-President, bade them welcome. The attendance was less than 20 per cent of the Fellowship. There was no council meeting due to the lack of a quorum, and apart from the President, the only other officer there was the Secretary. The meeting was in truth a forum set for tragedy.

Completing the duties of the first day, Albert Smith retired for 24 hours, and it is not stated who chaired the sessions on the second day, but an evening business meeting was handled by William Byford. Albert Smith finally appeared the morning of the third day and delivered his address. Following this, and on motion of Thaddeus Reamey of Cincinnati, "a vote of thanks was passed for the address, and for the manner in which the President had presided during those portions of the meeting that his condition had allowed of his occupancy of the chair." Then he took part in a discussion of a Fellow's paper and retired from the last meeting of the Society that he would ever attend. The newly elected President, William T. Howard of Baltimore, then took the chair and adjourned the ninth annual meeting, following the final paper of a session that must have lived in the memories of those present for a long time to come.

A few months after Chicago, and following a prolonged session in the operating room, Smith realized just before Christmas that he could not go on. A patient, in recording impressions of him, felt that his influence was due in great part to the sympathy and kindly interest extended to those whose troubles he took to himself without impatience. "I saw him," said she, "on the very day that he gave up, yet he was so bright and cheerful that I felt completely reassured that he was restored to health." He was invalided at home, and the advent of warm weather brought his removal to Beach Haven. This became a public task in a way, so beloved was he. It is recorded that municipal facilities provided for his removal to the train, through streets cleared by the police corps. Bedfast to some extent, and more so after the return to Philadelphia in the fall, he kept himself interestedly busy without complaint, and with gentleness and consideration for those about him. He read much, corresponded, and translated articles (for he had mastered German early—often studying during the odd hours of a home delivery). Continued sensitivity to the world about him and occupation with intellectual pursuits continued almost to the hour of his death on Dec. 14, 1885, in his fiftieth year. Whether or not he had requested one, an autopsy was performed. The destructive adenoma of the prostate gland was confirmed; malignancy in the renal tract had claimed the son as it had the father.

The funeral was held at the family residence, 1419 Walnut Street, a site long since replaced by a banking house. The rector of old St. Mark's Episcopal Church was present, along with well-known members of the Society of Friends, at the brief service attended by Smith's nearest professional and lay friends. A quotation from the "Public Ledger" a day or so later speaks for itself: "There were also present a number of young persons to whom the deceased had, by his gentle nature, endeared himself."

Albert Smith had not lived to see the victory of the causes he fought for to the end of his days, but 3 years afterward, on Jan. 20, 1888, the first woman physician, Dr. Mary Willets, was elected to membership in the Philadelphia County Medical Society. One has the feeling, however, that Smith must have sensed the crumbling opposition, and must have known that bit by bit the cause to which he had given so much of his strength would eventually be victorious.

It should be known also that it was not only for his efforts in the advancement of the medical education of woman that he shone forth. Colleagues told of his kindness and generosity to the underprivileged and to those in trouble. He appeared in court on occasion to help right considered injustices. His exalted conception of womanhood was evident in his personal association with patients and is recorded in an exceptional address he presented at the memorial service held at the Woman's Medical College on the occasion of the untimely death of Emmeline Cleveland, whose career has been mentioned earlier in this paper.

He wrote relatively few formal papers, but they were well prepared and laboriously worked over. Such was his final literary effort, his Presidential Address entitled "The Present Aspect of the Puerperal Diseases." In this treatise, testing all possibilities, he reviewed and discussed minutely the opposing views of Gaillard Thomas, who held the "absorptionist" viewpoint, and Fordyce Barker, who supported the "essential fever" theory. This debate had gone on in the New York Academy of Medicine during the winter of 1883-1884, creating widespread attention.

Years later, in 1914, J. Whitridge Williams and, in 1939, Frederick C. Holden, presidential essayists, criticized Albert Smith severely for denying and combating the bacterial origin and doctrine of the germ theory. Their contention was that a year before Albert Smith's presidential address, Garrigues and others of this Society were utilizing aseptic and antiseptic methods to reduce puerperal mortality. *Ipsa facto* this accusation is true, but intimate reading of Smith's

paper indicates that both critics may well have failed to grasp the true import of his objectivity. To my mind his comments were made in the spirit of fairness and truth characteristic of him. He was simply not ready at that moment to accept as an acknowledged fact that germs per se were the direct cause of puerperal sepsis. His discussion is replete with impartial comments on the views of believers, as it is with the comments of the nonbelievers. Referring to Pasteur he wrote: "When any one by repeated experiment proves the exact nature of the *causa morbi*, present in every case, and never found except under conditions of similar character, then I will accept the *corpus delicti*, even if it be Pasteur's diplococcus and 'microbes en chapelet' and honor him the more for his pertinacity in maintaining his ground."

Contrariwise, however, he cited the need for reliance upon Koch's postulates for definitive proof of the germ origin of puerperal fever. He did not categorically deny it, for he said, "I will not say that it is not true; I will not say that it may not be some day clearly proved."

Throughout his brief career Albert Smith had consistently stressed cleanliness and antiseptic obstetrics in his teaching, discussions, and formal papers. The procedures in labor that he mentioned in his address in 1884 were similar to those that the enlightened and justly praised Fellow of our Society, Henry Garrigues, strenuously put into effect in the fall of 1883, when he took over his tour of duty at the New York Maternity Hospital. During the preceding 9 months the mortality and morbidity figures in this hospital had been alarmingly high. With the enforcement of scrupulous cleanliness in all areas, plus chemical antiseptics, the morbidity was reduced and there were no deaths. As recollected, the term "asepsis" was not mentioned in Brooke Wells's touching memoir of Garrigues, which was the source consulted. Even though Albert Smith did not accept the germ theory, as his critics stressed, he rigidly practiced punctilious cleanliness and antiseptics in all the details at his command. He stood firmly as a scientist, seeking proof before acceptance. With this comment we bring to a close the discussion of Albert Smith's controversial address.

In the final year of his life, Albert Smith cherished the Honorary Membership awarded him by the Gynecological Society of Great Britain, among whose members he had cordial friends. He had long been Consulting Accoucheur to the Preston Retreat of ancient vintage. He had given of his energy and sympathy as Physician to the House of Refuge, forbiddingly named for its unfortunate occupants. The Hospital of the Good Shepherd in the Radnor countryside claimed him as Consulting Physician. Briefly during his early medical days he was an Attending Physician at the Philadelphia Hospital beyond the Schuylkill River, better known to some as "Blockley." For some years he served on the Board of Managers of the Wills Ophthalmic Hospital.

Classically educated and culturally inclined, Albert Holmes Smith, in company with other physicians of his day, became a member of the American Philosophical Society, the oldest learned society in the United States. Actually, the complete title is the "American Philosophical Society Held at Philadelphia for Promoting Useful Knowledge." Its origin stems from 1727, and in 1769 Benjamin Franklin became its first President when it combined with a society Franklin had formed years before—the "American Society for Promoting Useful Knowledge."

Albert Smith's membership in the Philosophical Society brought him in closer association with a singular and brilliant person. Harrison Allen was a renowned anatomist with both medical and dental degrees. A scientist of high order, he was competent in comparative anatomy, zoology, and anthropology, and he had held various chairs at the University of Pennsylvania. Harrison Allen was requested by the American Philosophical Society to prepare the memoir of Albert Holmes Smith. I am thankful to have found this reference,

for I can conceive of no more fitting way by which to emphasize Smith's story than by quoting excerpts from Harrison Allen's precise and discerning tribute:

Doctor Smith was *facile princeps* the leading obstetrician of his time in America. He developed a skill in the management of cases requiring instrumental aid which in the opinion of his colleagues has probably never been excelled. . . . It has been frequently observed that men of the cast of mind of Dr. Smith are rarely literary in their inclinations. . . . It is a noteworthy circumstance that in spite of this disinclination Dr. Smith was a literary worker . . . he was always self-depreciatory of his literary abilities and disclaimed every intention to authorship. . . . It was more to him to right a wrong and to defend the weak than to make a scientific discovery. . . . He was not heard to express any exalted conception of the duties of the physician; he made no pretention to any special consecration to good works.

Albert Smith was not forgotten by his colleagues in the Society. At the meeting in Washington the next fall (1885), Joseph Tabor Johnson presented a resolution to be sent Smith tendering "a vote of sympathy in his affliction, with the hope of his speedy restoration to health." In seconding this, Henry P. C. Wilson of Baltimore, in the formal language of that day, stated that he was pleased to do so, "because I regard Albert H. Smith as one of the ablest and most charming men we have in our Society. I think it eminently proper that we should take this action and feel very much obliged to Dr. Johnson for presenting these resolutions which may assure Dr. Smith in his suffering that he has not passed entirely out of our thoughts."

Courageous in death, as he was in life, Albert Holmes Smith remained steadfast in misfortune, uncomplaining and serene. I trust that you, who have listened so patiently, can understand why I became so deeply interested in this gentleman, and why I chose the subject, "Albert Holmes Smith: A Study in Courage."

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AN IMMUNOLOGIC CHARACTERISTIC OF THE SERUM OF NORMAL PREGNANCY*†

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SINCE the time of Obata,¹ many investigators have sought to explain certain disorders of pregnancy on an immunologic basis, including allergic reactions to substances of placental origin.² This report concerns an attempt to re-explore this aspect of pregnancy and to examine by newer immunologic means the sera of pregnant women for the presence of a substance or substances peculiar to pregnancy.

A number of methods are now available to permit a visible separation of antigens by allowing them to react in a gel with their specific, precipitating antibodies. This physical separation is the result of the antigens' diffusing through the gel at different rates. The Ouchterlony method of precipitin analysis³ and its modifications⁴ have been chosen because they have certain advantages over other methods. First, a visible line appears in the gel when an antigen and its specific antibody precipitate. Second, it is possible to demonstrate the presence of a particular antigen in a complex mixture. Third, the possible similarity between antigens may be illustrated; this concerns itself with the problem of partial identity which will not be pursued here.

Materials and Methods

The following antigens were utilized in the experiments: PS—serum from normal pregnant women; NS—serum from normal nonpregnant women; CG—chorionic gonadotropin (Squibb).

The normal pregnancy serum was obtained from patients in the third trimester of pregnancy. Women with no previous history of pregnancy supplied the normal nonpregnancy serum.

The antibody was Anti-PS—antiserum against the serum of normal pregnant women. The antiserum (Anti-PS) was produced in rabbits against the pooled serum of 6 patients in the third trimester of normal pregnancy. The animals were injected intravenously with 0.25 ml. of serum every other day for

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22 days. Five to 7 days after the last injection the rabbits were bled by cardiac puncture. The various sera were frozen until they were utilized; then they were thawed and stored in the refrigerator at 3° C.

The diffusion plates were Petri dishes 10 cm. in diameter containing a 1 per cent agar gel made with distilled water (pH 7.4).⁵ Three wells, produced in the gel by metal molds which were removed after hardening, served as reservoirs for the reactants in the serologic analysis. The plates were stored in the refrigerator at 3° C. at all times; this was sufficient to preserve asepsis.

The experiments were performed by use of duplicate plates, and the results reported are based upon identical findings on both plates. Demonstration of the reaction was dependent upon a satisfactory antiserum, the quality of which varied with different lots. When the results of a duplicate set differed, the plates were discarded.

The number of lines formed, and their position with respect to the wells, depended upon the number of antigens and antibodies present, their concentrations, and the diffusion coefficients of each, the latter being determined by the physical characteristics of the substance. The "precipitation spectrum" is illustrated diagrammatically in Fig. 1, with the hypothetical antigens and antibodies designated *a*, *b*, and *c*, and *A*, *B*, and *C*, respectively.

To simplify the identification of unknown antigens, the techniques of simultaneous inhibition,⁶ preinhibition⁴ and specific antibody absorption⁷ were

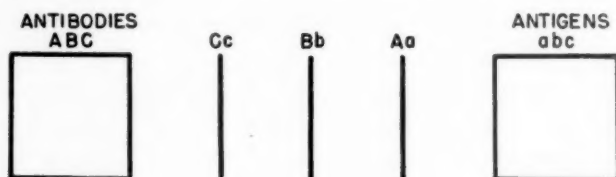


Fig. 1.—Ouchterlony precipitation spectrum.

employed. These techniques prevent certain antibodies from participating in the serologic analysis. Thus, a specific antiserum can be used in the plate to identify a particular antigen in a test serum.

Simultaneous inhibition and preinhibition are based on specific antibody absorption, a procedure by which the appropriate serum and antiserum are added to the well or wells for antibody absorption. The essential differences in the details of the two methods are considered in the following discussion and are illustrated in Figs. 2 and 3.

In simultaneous inhibition (Fig. 2) the nature of the antibody or antibodies diffusing from the inhibited well can be controlled by using amounts of serum and antiserum optimal for antibody absorption.⁸ This type of inhibition is performed by adding to a single well a serum containing antigens *a* and *b* and an antiserum containing antibodies *A*, *B*, and *C*. The unmatched antibody (*C*) is unaffected and diffuses freely into the gel. Hence the only line of precipitation which appears between the upper left well and the lower well is formed by the reaction of the antibody (*C*) and its matching antigen (*c*) in the test serum.

In the preinhibition technique (Fig. 3) the diffusion plate is saturated with appropriate antigens (*a* and *b*) prior to the actual analysis in order that their corresponding antibodies (*A* and *B*) will be precipitated, either at the edge of or within the wells, during the experiment. It is very necessary, therefore, that the amounts of both the preinhibiting serum (*a* and *b*) and the antiserum (*A*, *B*, and *C*) conform to the optimal ratio for antibody absorption. When the test serum (*a*, *b*, and *c*) and its antiserum (*A*, *B*, and *C*) are added to the designated

wells, only unknown antigens and their matching antibodies will be available for reaction. Hence, the only line of precipitation that appears is formed by the reaction between the antigen (*c*) and its antibody (*C*).

By the method of specific antibody absorption,⁷ an antiserum was prepared which contained only those antibodies corresponding to the antigens peculiar to PS and was free of precipitated antigen-antibody complexes. This was done by mixing optimal proportions of NS and Anti-PS in a test tube, incubating the mixture in the refrigerator for 24 hours, and then removing the precipitate by centrifugation.

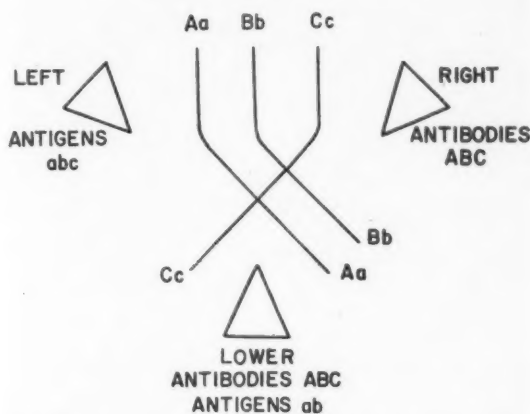


Fig. 2.—Modification of antigen analysis (simultaneous inhibition).

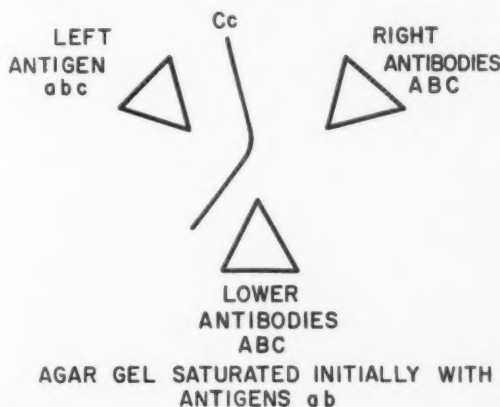


Fig. 3.—Björklund modification of antigen analysis (preinhibition).

Results

Investigation has revealed a serologic difference between normal pregnancy serum (PS) and normal nonpregnancy serum (NS). The substance or substances responsible for the serologic distinction of normal pregnancy serum is antigenic in the rabbit and is here demonstrated by the Ouchterlony method of serologic analysis.

Demonstration of a Serologic Characteristic of Normal Pregnancy by Simultaneous Inhibition.—The design of the experiment is indicated in Fig. 4. The lower well in Fig. 4, A was inhibited with normal nonpregnancy serum (NS), while the lower well in Fig. 4, B was inhibited with normal pregnancy serum (PS). These were the only wells inhibited.

The upper wells containing PS and Anti-PS were filled initially with 0.05 ml. of reactant and 0.05 ml. of normal saline while the lower wells were filled with 0.05 ml. of Anti-PS, 0.025 ml. of NS or PS, and 0.025 ml. of normal saline. This ratio between serum and antiserum in the lower wells conformed to the optimal ratio for complete absorption of antibodies. The wells were refilled on the second, fourth, and sixth days of incubation and photographs were prepared on the tenth day.

A.



B.

Fig. 4.—Demonstration by simultaneous inhibition. *A*, Experimental. Upper left well—PS; upper right well—Anti-PS; lower well—Anti-PS and NS. *B*, Control. Upper left well—PS; upper right well—Anti-PS; lower well—Anti-PS and PS.

In Fig. 4, *A* the line arising from the apex of the precipitation spectrum and continuing as a straight line between the upper left well and the lower well was considered to be significant, in view of the absence of a similar line on Fig. 4, *B*. Inasmuch as this line occurred in Fig. 4, *A*, inhibited with NS, and failed to appear in Fig. 4, *B*, inhibited with PS, it was considered to be evidence of a substance peculiar to normal pregnancy serum.

Demonstration of a Serologic Characteristic of Normal Pregnancy by Preinhibition.—An experiment employing preinhibition was performed to obtain

additional evidence for the existence of a substance or substances specific to normal pregnancy serum. The design of the experiment is indicated in Fig. 5.

The technique of preinhibition comprised two separate steps. First, the serum (NS or PS) used for inhibition was allowed to diffuse into the gel before the analysis began, as implied by the name of the technique. Second, the anti-serum (Anti-PS) was added to the wells at the same time that other reactants were added to their respective wells for the serologic analysis.

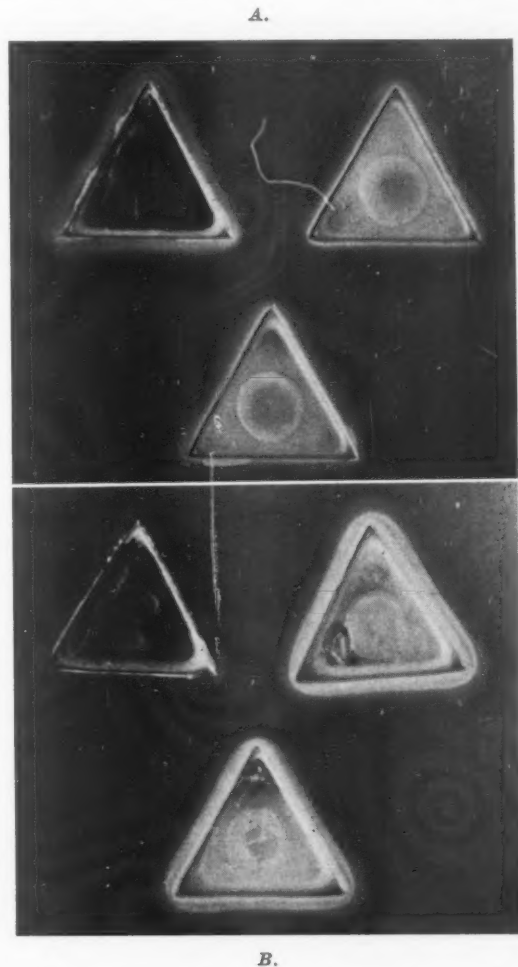


Fig. 5.—Demonstration by preinhibition. *A*, Experimental. Upper left well—PS; upper right well—Anti-PS; lower well—Anti-PS. Plate preinhibited with NS. *B*, Control. Upper left well, PS; upper right well—Anti-PS; lower well—Anti-PS. Plate preinhibited with PS.

Preinhibition of the plate in Fig. 5, *A* was effected by adding 0.05 ml. of NS and 0.05 ml. of normal saline to each of the three wells. The plate in Fig. 5, *B* was preinhibited with PS in a similar manner. After the wells were empty, which required 3 or 4 days, the plates were used for the serologic analysis.

The analysis was begun by filling each well of both plates with 0.05 ml. of the indicated reactant and 0.05 ml. of normal saline. On the second and fourth days of incubation the wells were refilled, and on the tenth day the plates were photographed. The optimal ratio between the preinhibiting serum (NS or PS)

and the antiserum (Anti-PS) for maximum antibody absorption in each plate was attained by using a total volume of preinhibiting serum of 0.15 ml. and a total volume of antiserum of 0.30 ml.

The presence of two lines of reaction on the plate in Fig. 5 A indicates that inhibition of the antiserum (Anti-PS) by NS was incomplete. Two antibodies, unabsorbed by NS, diffused from the wells containing Anti-PS and reacted with antigens present in normal pregnancy serum. However, when PS was used to inhibit (Fig. 5, B), the lines did not appear and their absence indicated that all of the antibodies in Anti-PS were absorbed by PS. The results of this experiment are interpreted as evidence of a specific serologic characteristic of normal pregnancy serum.

Demonstration of a Serologic Characteristic of Normal Pregnancy Serum by Specific Antibody Absorption.—The design of the experiment is illustrated in Fig. 6. An anti-serum specific for those antigens peculiar to normal pregnancy serum (PS) was prepared by absorbing Anti-PS with NS, on the basis of the

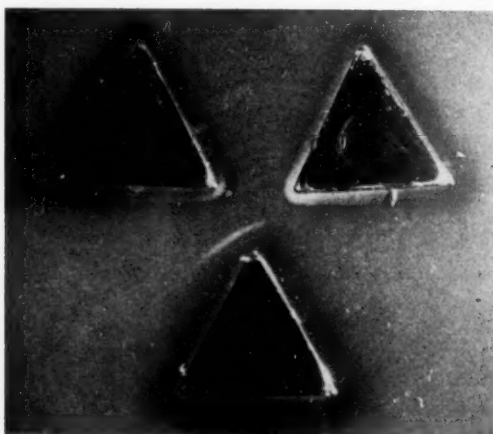


Fig. 6.—Demonstration by a specific antiserum. Upper left well—PS; upper right well—NS; lower well—Anti-PS absorbed with NS.

optimal ratio, and removing the precipitate by centrifugation. Unlike the procedures of simultaneous inhibition and preinhibition, this specific antiserum was prepared in a test tube prior to its use in the plate.

Initially, PS and NS were added to the upper wells in 0.05 ml. amounts together with equal volumes of normal saline. To the lower well was added 0.1 ml. of specifically absorbed antiserum. On the fourth day the wells were refilled, and the plates were photographed on the sixth day of incubation.

The presence of two straight lines on the field between the PS and the Anti-PS wells, being unmatched on the NS side of the field, are interpreted as two antigens peculiar to PS. This serologic distinction of PS is a qualitative characteristic within the sensitivity of the method of precipitin analysis.

In an experiment of the type illustrated in Fig. 6 chorionic gonadotropin was substituted for NS in the upper right well to test its identity with one of the two antigens shown to be peculiar to normal pregnancy serum. During the experiment chorionic gonadotropin failed to give rise to a line of reaction under conditions favorable for the demonstration of the pregnancy antigens.

The possibility was considered that the serologic characteristic of pregnancy serum might be due to either the A or B blood group isoagglutinin. To test this possibility the sera of 6 pregnant women of blood group O were absorbed with AB erythrocytes to remove the respective isoagglutinins, and then tested with

Anti-PS, which had been absorbed with nonpregnancy serum. It was possible to demonstrate the presence of the antigens associated with pregnancy in each of the 6 sera after the absorption of isoagglutinins.

The serologic reaction characteristic of normal pregnancy was found to occur in each of 29 sera obtained from patients during the last trimester of pregnancy.

Summary

An immunospecific characteristic of normal pregnancy serum has been demonstrated through the application of the Ouchterlony-Björklund serologic method. While the three techniques gave rise to different results, one line of precipitation with simultaneous inhibition and two lines of precipitation both by preinhibition and by specific antibody absorption, the results may not be so dissimilar as they first appear. In simultaneous inhibitions, the obvious conclusion that the single line of reaction means a single antigen may not be valid since a known limitation of gel analysis may be the reaction of two similar antigens to form a single line of precipitation.³ Hence, the presence of two antigenic substances is not excluded by the evidence of a single line of precipitation. In preinhibition, the two discrete lines of precipitation should probably be interpreted as evidence of the presence of at least two antigenic substances in the serum of normal pregnancy. However, a single antigenic substance reacting to form two discrete lines has been acknowledged as a possibility in this method.³ Thus, the evidence gained by the three methods of antibody absorption is interpreted to mean the presence of at least one and perhaps two substances peculiar to the serum of normal pregnancy which, by virtue of their antigenicity in the rabbit, can be demonstrated by serologic methods. Considering the lack of reactivity of chorionic gonadotropin in the gel, it is unlikely that it is one of the antigenic substances.

Further, the reaction is not due to a blood group A or B isoagglutinin. Thus the identification and the significance of the substance or substances which have been demonstrated in pregnancy serum remain to be established. A considerable amount of speculation could be indulged in concerning fetomaternal isoimmunization. However, on the basis of our present evidence, we would prefer to defer making any such remarks until we have made more progress in the identification of these antigens and studied their relation to pregnancy.

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Discussion

DR. CURTIS J. LUND, Rochester, N. Y.—The search for immune substances in the serum of women with disorders of pregnancy is an old one. Some results were reported,

others were not. These older studies bogged down for several reasons, but an important one was the inadequacy of the test tube. When antigen and antibody are united in solution, a precipitate forms quickly so study of any part of the completed reaction is impossible.

The Ouchterlony principle has provided a comparatively new, simple qualitative test for separating antigens. The technique as applied in this study is an accepted experimental method. A few minor questions about the experimental procedure might be raised, however:

1. The immunized rabbits were bled within 5 to 7 days after the last injection of PS. In this comparatively short time is it not possible that some of the antigen is still present in the rabbit's circulating blood?

2. The authors have mentioned rejection of results whenever duplicate plates differed. It would be of interest to know the nature and frequency of this disparity.

3. To obtain the characteristic serologic reaction it was necessary to have a "satisfactory antiserum" produced by the rabbit. Variations in the quality of these antisera were observed. What techniques were used to exclude the antigen of the individual patient as a factor in the quality of the serologic reaction?

Dr. Reid and his associates have clearly demonstrated specific immune substances in the serum of pregnant women in the third trimester of pregnancy. These substances could be maternal antibodies similar to the "globulin antibody" of rabbits used in the Coombs test. They could be fetal substances which are circulating in the maternal blood or they might be placental products. Pregnanediol is not usually considered to be antigenic. However, in our laboratory Heckel has demonstrated clinical sensitivity to unconjugated pregnanediol. Support of this concept has been provided by the recent work of Beiser and associates,¹ who suggested that progesterone and other steroid hormones may act as haptens when coupled with a protein. They were able to produce specific antibodies in rabbits by injection of steroid-protein conjugates.

Reference

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DR. REID (Closing).—The answer to the question concerning inconstant results resides in the production of an antisera with a sufficiently high titer, a difficulty encountered in preparing antisera generally. I am not in a position to furnish an answer as to the identity of these antigenic substances, but it is difficult to see where residual human antigens in the antisera would influence the results. I believe that hormones—steroid hormones particularly—are not antigenic.

Perhaps Dr. Douglas's paper to follow may furnish some evidence as to the source of this (these) antigenic substance(s). More interesting and, I am sure, more important is why the pregnant woman can tolerate the antigenic component of this genetically different new individual, and not reject this so-called homograft, the fetus, before the period of medical viability.

ABRUPTIO PLACENTAE AND RUPTURE OF THE MARGINAL SINUS OF THE PLACENTA: SOME RELATIONSHIPS*

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THE purpose of this report is to present some similarities and dissimilarities of abruptio placentae and rupture of the marginal sinus of the placenta. It is hoped that these observations will lead to a better understanding of each of these complications. To make contrast between these 2 placental accidents we have drawn on a 3½ year experience at the University of Miami-Jackson Memorial Hospital with 113 cases of abruptio placentae and 130 cases of rupture of the marginal sinus.

Diagnosis

Anatomic criteria for the diagnosis of abruptio placentae and rupture of the marginal sinus were demanded in the material reported here. No diagnosis of abruptio placentae or rupture of the marginal sinus was made solely on the clinical course of the case. If a placenta under consideration did not show unmistakable evidence of either of these lesions the diagnosis was listed in the hospital records as "Cause Undetermined," unless, of course, there was some other undoubted diagnosis. Practically all of the placentas were examined by the senior author as soon after delivery as convenient. The appearance of the placenta at the time of delivery was discussed with the patient's attendants.

Abruptio Placentae.—All of the 113 cases diagnosed as abruptio placentae showed changes on the maternal aspect of the placenta that were considered characteristic of this lesion. The placenta exhibited one or more of the following signs: a sizable clot attached to the maternal surface, a depression obviously caused by a clot, or unequivocal changes in the color and texture of the maternal surface which represent abruptio placentae. (Fig. 1 is a typical example of this type of placenta.)

Rupture of the Marginal Sinus.—All of the 130 cases of rupture of the marginal sinus which form the basis for this report provided placentas which satisfied our anatomic criteria for the diagnosis. An attendant at delivery noted that there was a clot at the margin of the placenta and that that clot was continuous with clotted blood in the marginal sinus. If examination of the placenta indicated that the clot had ever been between the maternal surface of the placenta and the uterus, the diagnosis of abruptio placentae was made and not that of rupture of the marginal sinus. The diagnosis of placenta previa

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was eliminated by vaginal examination at some time unless this examination was precluded by some obvious circumstance such as an imminent delivery. Figs. 2, 3, and 4 show typical specimens of rupture of the marginal sinus.

Observations on Placentas in Rupture of the Marginal Sinus

In preparation for a description of certain similarities of the placentas in abruptio placentae and rupture of the marginal sinus; it will be necessary to record some observations we have made in cases of rupture of the marginal sinus.

A. Observations on Surface Area of the Placentas.—This portion of the observations is a negative report but it is necessary to introduce it in order to identify the control placentas which are important in observations that follow.

Earlier in our investigations of rupture of the marginal sinus there was reason to suspect that the placentas with rupture tended to be large placentas—

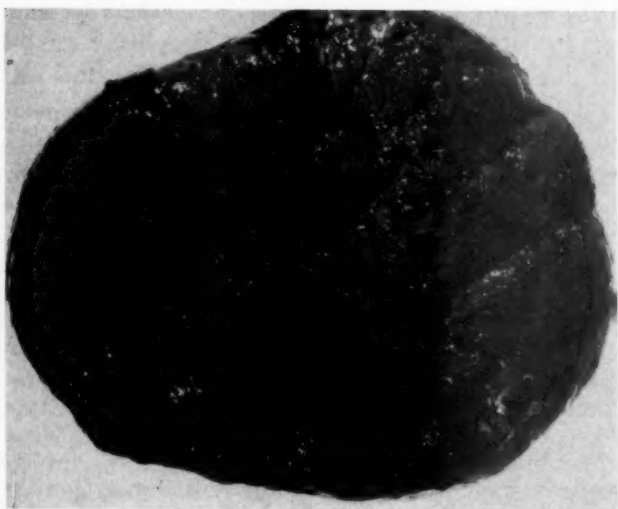


Fig. 1.—Abruptio placentae, showing, on the left, the typical change in color and general appearance of the separated portion.

that is, the surface areas of the placentas were greater than usual. This impression was not supported as larger number of cases of rupture of the marginal sinus became available for study. In order to test the validity of this initial impression, an effort was made to study some random placentas with rupture of the marginal sinus and some suitable controls. The random placentas, from cases of rupture of the marginal sinus, were laid flat on paper and a tracing was made of the periphery of the placenta. Also, 56 random control placentas from pregnancies of matching weeks of gestation were collected. With the exception of cases of premature labor, the control placentas came from normal pregnancies. Tracings were also made of these control placentas. The surface areas of all the tracings were measured with a planimeter. These surface areas were carefully compared and there were no significant differences between the control placentas and the placentas with rupture of the marginal sinus.

B. Inferior Position on the Placenta of the Ruptured Sinus.—Early in these studies we realized that the rupture of the marginal sinus had usually occurred at the edge of the placenta that was lowermost inside the uterus. We do not claim this is an original observation.

The edge that had been lowest inside the uterus can be determined by examining the membranes and the placenta. The rent in the membranes represents the location of the cervical opening; therefore, the placental edge nearest the rent must have been the dependent edge in the uterus. Except when there

Fig. 2.

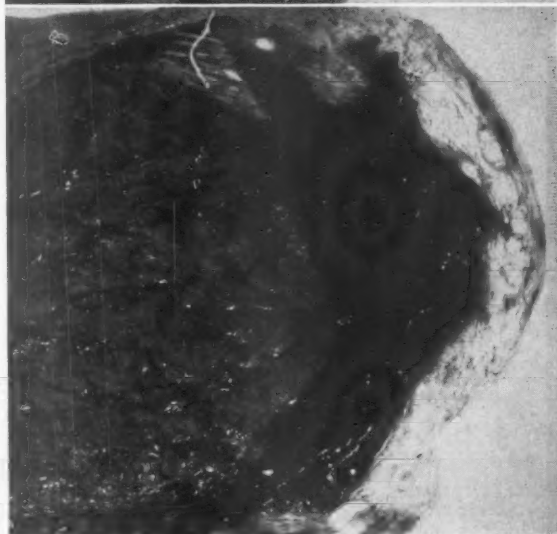
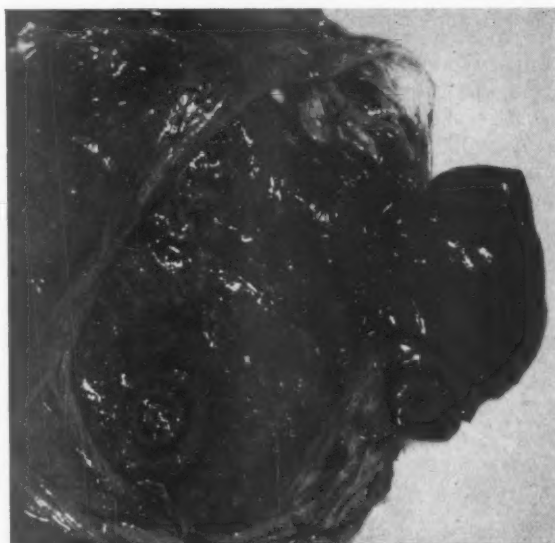


Fig. 3.

Figs. 2 and 3.—Typical examples of rupture of the marginal sinus.

has been a direct fundal position of the placenta, this rent in the sac will be found in an eccentric position, which permits the examiner to note quite definitely which edge has been inferior.

To increase the significance of this observation we are adding the 51 cases of rupture of the marginal sinus that the senior author studied on the Tulane Service at the Charity Hospital at New Orleans to the 130 cases found at the

Jackson Memorial Hospital.¹ In 51 of the total of 181 cases, the membranes-ruptured sinus-placenta relationship was either not sought or the specimen was too damaged by handling to be of value. In 130 cases, the positions of the rent in the membranes and the ruptured marginal sinus were observed and recorded. In 6, there had been a central fundal location of the placenta, and no portion of the edge of the placenta could have been lower in the uterus than any other portion of the edge. In 123 of the 130 cases suitable for this observation, it was noted that the rupture of the marginal sinus had occurred at the lowermost edge of the placenta. In only one instance was the ruptured sinus on the superior half of the placenta. We interpret these observations as meaning that the edge of the placenta lowest in the uterus is the area where the marginal sinus is most vulnerable to rupture. Presumably, the marginal sinus in this area can be opened by the formation of the lower uterine segment, dilatation and effacement of the cervix, or traction of the presenting part of the fetus on the membranes. But we cannot eliminate the possibility that the sequence of events may be the reverse—that is, that the rupture of the marginal sinus caused or accelerated the changes in the uterus.



Fig. 4.—Typical example of rupture of the marginal sinus.

C. Low Implantation.—While studying the earlier cases of rupture of the marginal sinus, the senior author had the impression that often in this syndrome a placental edge had been relatively close to the cervical os. He suspected this proximity because, in many cases, the membranes were extremely short on the side where the rupture of the marginal sinus had taken place. In some cases, there were no membranes, or nearly none, attached to the placental edge, which means one side of the opening into the sac was formed by the placental edge. The closer the placenta is to the cervical os, the shorter the membranes should be that are attached to the lower edge of the placenta.

In these cases from the Jackson Memorial Hospital we have attempted to verify this impression by measuring the length of the membrane adjacent to the ruptured marginal sinus. In 126 of the 130 cases, we were able to record this length of membranes which represents the distance of the placenta from the cervical os. In 43 of these 126 cases, the membrane length was 2 cm. or less. In 13 cases the membrane length was recorded as 0 cm. This measurement was possible in 31 of the 56 control cases mentioned in Paragraph A, and only 3

of these 31 control placentas had membrane lengths that were 2 cm. or less. The difference of the membrane lengths in these two groups can be seen in Fig. 5.

A chi-square significance test applied to these data indicates that a difference in membrane length, marked as that observed between the normal group and the rupture of the marginal sinus group, would occur by chance less than once in a thousand times ($p < .001$). The observed difference is thus considered to be of high statistical significance.

Our interpretation of these observations is the rather obvious one that in the rupture of the marginal sinus syndrome an edge of the placenta often has a lower position in the uterus than usual, and this low implantation abets the tearing of the marginal sinus in the dependent portion of the placenta.

While the placenta was implanted low in the uterus in many of the cases of rupture of the marginal sinus, we should point out that the placental edge could not be palpated during vaginal examination in any of these cases. In

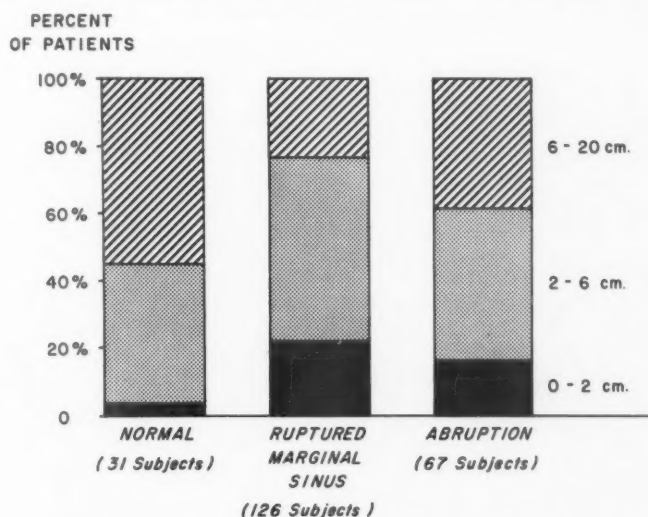


Fig. 5.—A comparison of the membrane lengths (distance from inferior edge of the placenta to cervical os) in normal patients and in patients with ruptured marginal sinus or abruptio placentae.

the years covered by this study, there were a few cases in which a placental edge was palpated but did not overlie the internal cervical os. These cases were designated "low implantation."

Similarities of the Placentas in Cases of Abruptio Placentae and Rupture of the Marginal Sinus

A. Coexistence of Abruptio Placentae and Rupture of the Marginal Sinus.—In examining the placentas of some patients with abruptio placentae, we noted that the retroplacental clots were continuous with clotted blood in the marginal sinus of the placenta. The important difference between these special cases and the rupture of the marginal sinus syndrome is that, in rupture of the marginal sinus, the clot is at the margin of the placenta and there is no evidence that the clot was ever in a retroplacental position.

All of the placentas with abruptio placentae were not examined for these coexisting lesions because we were not aware of this variety until relatively recently. In 21 of the 113 cases of abruptio placentae at the Jackson Memorial Hospital, this combination of abruptio placentae and rupture of the marginal

sinus was found. It was thought best to consider these cases as abruptio placentae since that was the more important or more dangerous of the 2 lesions. Furthermore, the clinical picture of each case was that of abruptio placentae.

In the cases of coexisting abruptio placentae and rupture of the marginal sinus we have found no proof of which one precedes the other—if one does precede the other rather than the 2 occur simultaneously.

B. Location of Separated Portion of the Placenta in Partial Abruptio Placentae.—In partial abruptio placentae, we noted a tendency for the separated portion of the placenta to be the portion that had been the lowermost part of the placenta inside the uterus. This position of the placenta was determined by noting the relation of the separated portion of the placenta to the rent in the membranes as described in Paragraph *B* above.

In 63 of the 113 cases of abruptio placentae at the Jackson Memorial Hospital, observations on this point could not be made for various reasons, such as: complete separation of the placenta; distortion of the placenta by manual removal or by handling at cesarean section; central location of the placenta in the fundus; or incomplete examination of the placenta and membranes immediately after delivery. In 50 cases of partial abruptio placentae accurate observation was possible. In 45 of these cases (40 per cent of all cases of abruptio placentae) the separation was partial and the separated portion of the placenta was on the same side as the rent in the membranes; that is, it was the portion of the placenta that had been dependent in the uterus. In only 4 cases, the separated segment was on the side farthest away from the rent, that is, the part that had been superior in the uterus. In one case, the rent was at the apex of the membranes and was of no value in this particular observation.

C. Low Implantation in Abruptio Placentae.—It has already been stated that the placenta that has a rupture of its marginal sinus is a placenta that often is implanted low in the uterus. The two observations above (Paragraphs *A* and *B*) on the placenta, after first being made and then confirmed in succeeding specimens, naturally led in time to a search to learn if there is anything unusual about the position of the separated placenta inside the uterus. As described earlier, the height of the placenta inside the uterus (the distance of the placental edge from the cervical os) can be determined by measuring the shortest portions of the membranes that are attached to the placenta. The length of this membrane was measured in 67 cases of abruptio placentae. More of these observations on the placenta were not obtained because of the unsuitability of some specimens and the relatively recent date this measurement was recognized as being important.

TABLE I. COMPARISON OF THE SHORTEST MEMBRANE LENGTHS IN PATIENTS WITH RUPTURE OF THE MARGINAL SINUS OR ABRUPTIO PLACENTAE AND IN NORMAL PATIENTS

SHORTEST LENGTH OF MEMBRANE (CM.)	NORMALS		RUPTURE OF THE MARGINAL SINUS		ABRUPTIO PLACENTAE	
	NO.	%	NO.	%	NO.	%
0 to 2	1	3	28	22	11	16
2 to 6	13	42	69	55	30	45
6 to 20	17	55	29	23	26	39
Total	31	100	126	100	67	100

Table I shows the distribution of the membrane lengths for each of three groups of patients—normal, abruptio placentae, and rupture of the marginal sinus. The normal patients are the 31 cited as control patients in the section of this paper dealing with the surface area of the placentas in rupture of the

marginal sinus. The 126 patients with rupture of the marginal sinus and the 67 patients with abruptio placentae are those described earlier as providing specimens suitable for measuring this membrane length. Fig. 5 graphically represents the data given in Table I. As mentioned in the section dealing with the tendency to low implantation in rupture of the marginal sinus, there is a striking difference between the group of normal patients and the group with rupture of the marginal sinus. The group having abruptio placentae can be seen in Fig. 5 to be more like those with rupture of the marginal sinus than like the normals. This appears to support the impression that in abruptio placentae the placenta has a tendency toward low implantation.

These studies are being continued. We recognize from statistical treatment of the figures that it will be most profitable to enlarge the group of controls.

Comparison of the Clinical Problems in Abruptio Placentae and in Rupture of the Marginal Sinus

When cases of abruptio placentae and rupture of the marginal sinus were diagnosed by strict adherence to anatomic criteria, and only anatomic criteria, 2 distinct clinical syndromes emerged. Table II lists some of the important characteristics of the cases of abruptio placentae and rupture of the marginal sinus used in this report and serves to compare the frequency of these characteristics in the 2 syndromes.

TABLE II. CHARACTERISTICS OF ABRUPTIO PLACENTAE AND RUPTURE OF THE MARGINAL SINUS

	ABRUPTIO PLACENTAE	RUPTURE OF THE MARGINAL SINUS
No. of cases	113	130
Ratio to No. of deliveries	1:166	1:144
Incidence of premature babies	50%	46%
Incidence of vaginal bleeding	82%	100%
<i>Dissimilarities.—</i>		
Shock	11%	1%
Blood loss		
Median	600 ml.	200 ml.
Per cent of patients losing 1,000 ml. or more	26%	2%
Abdominal pain	45%	rare
Uterine tenderness	34%	rare
Uterine hypertonicity	57%	rare
Hypofibrinogenemia	20%	none
Toxemia	42%	17%
Stillbirths	55%	2%

There are no great disparities between abruptio placentae and rupture of the marginal sinus in the prevalence of the sign of vaginal bleeding or delivery of premature infants, or in the incidence of these lesions. Eighty-two per cent of the 113 women with abruptio placentae had external, or vaginal, bleeding. Although rupture of the marginal sinus can be present without external blood loss, there were no such instances in this study of 130 cases.

Fifty per cent of the 111 single births to the women with abruptio placentae were premature (2,500 grams or less). The 46 per cent premature rate in rupture of the marginal sinus represents one of the important characteristics of that syndrome and carries its only real threat to life. This marginal sinus premature group suffered chiefly from a high neonatal mortality rate. Fifty-four of the 58 premature babies were born alive, but 19 died

in the neonatal period. Seven of the 19 infants weighed less than 1,000 grams and 15 less than 1,500 grams. All of these 19 babies were born before the thirtieth week of gestation.

Fish and associates² reported that rupture of the marginal sinus was the second most common cause of premature labor, second only to premature rupture of the membranes.

The basic dissimilarity between abruptio placentae and rupture of the marginal sinus is in their danger to the mother and fetus. Abruptio placentae carries grave risks for both mother and fetus, while rupture of the marginal sinus poses less of a threat. Rupture of the marginal sinus as a diagnosis is important primarily because it can be mislabeled abruptio placentae; concepts of the latter and of the best way to manage it clinically are faulty when based on experiences with collections of cases that are not all abruptio placentae.

It is understandable that true abruptio placentae, with its dissolution of the placenta-uterine union and its shock, should have a high stillbirth rate. Despite a readiness to do cesarean section we had a 55 per cent stillbirth rate; fetal heart tones could not be heard on admission to the hospital in 43 per cent of these cases. The stillbirth rate among the women with rupture of the marginal sinus was only 2 per cent.

At the University of Miami we have been able to make the antepartum diagnosis of the 2 complications which are the subject of this paper with a reasonable accuracy because of the dissimilarity in the symptoms. Seventy-six per cent of the patients with abruptio placentae had one of the signs we so frequently see with it: uterine hypertonicity, abdominal pain, uterine tenderness, or shock (Table II). Thirty-two per cent had the complete triad of pain, hypertonicity, and tenderness of the uterus; these signs were rarely noted in cases of rupture of the marginal sinus and then generally appeared in low intensity.

Evidence of our success in the antepartum diagnosis of abruptio placentae, and hence in the predominance of symptoms, is seen in our cesarean section rates. We have described our management of abruptio placentae as aggressive, and this frequently means cesarean section. Our management of suspected rupture of the marginal sinus is expectant. The 113 patients with abruptio placentae in the 3½ year study period had a cesarean section rate of 32 per cent and no sections were done in the 130 cases of rupture of the marginal sinus.

The difference between abruptio placentae and rupture of the marginal sinus in the amount of blood loss accounts in part for the greater frequency of shock in the former (11 per cent compared to 1 per cent). The median estimated blood loss in the cases of abruptio placentae was 600 ml., and only 200 ml. in cases of rupture of the marginal sinus. Twenty-six per cent of the patients with abruptio placentae lost 1,000 ml. or more of blood, whereas this degree of blood loss was present in only 2 per cent of the patients with rupture of the marginal sinus.

Defect in blood clotting was a major problem in the cases of abruptio placentae. Although not so zealously sought for, no clotting abnormalities were observed in our cases of rupture of the marginal sinus. In patients suspected of having abruptio placentae, we attempted to record in as many cases as possible, and we repeated as frequently as indicated, the following 4 observations: quantitative fibrinogen levels; qualitative fibrinogen tests (Fibrindex); test tube clot observation tests; and signs of abnormal bleeding tendencies. Our observations on 113 cases of abruptio placentae can be summarized by stating 20 per cent of these patients had quantitative fibrinogen

levels below 150 mg. per cent; 32 per cent had one or more abnormalities among the 4 types of observations mentioned above; 12 patients exhibited hemorrhagic diathesis; and 21 per cent needed intravenous fibrinogen therapy.

A fundamental difference between abruptio placentae and rupture of the marginal sinus is in the prevalence of toxemia. Forty-seven (42 per cent) of the patients with abruptio placentae had toxemia of pregnancy. Two of these women had eclampsia. We use the word "toxemia" in a general sense, including all hypertensive disease found in pregnancy. Nelson and Brewer,³ in a survey of the staff patients at the Jackson Memorial Hospital, used the same criteria for the diagnosis of toxemia as we used in this study of abruptio placentae and rupture of the marginal sinus. They found a toxemia incidence of 14.4 per cent. The incidence of toxemia among our patients with rupture of the marginal sinus was only 17 per cent—practically the same as the hospital incidence.

This wide difference in the incidence of toxemia between cases of abruptio placentae and rupture of the marginal sinus, plus the fact that this difference is the only recognized dissimilarity that pre-existed the onset of these placental accidents, suggest that toxemia may be a factor that determines whether or not there is to be a rupture of the marginal sinus or abruptio placentae.

Placenta Previa

Placenta previa is not the topic of this paper, nor was placenta previa as carefully analyzed as abruptio placentae and rupture of the marginal sinus during the 3½ year study period. However, certain similarities between placenta previa and rupture of the marginal sinus should be mentioned. First, placenta previa is a hemorrhagic complication of pregnancy and frequently the major source of the bleeding is the marginal sinus of the placenta. Some specimens of marginal placenta previa have the same appearance as typical specimens of rupture of the marginal sinus of the normally located placenta; there can be a clot at the placental margin and that clot can be continuous with clotted blood in the marginal sinus.

Placenta previa, of course, has the low implantation characteristic of rupture of the marginal sinus. We have suggested that this may also exist in a significant number of cases of abruptio placentae. In symptomatology, rupture of the marginal sinus resembles marginal placenta previa more than anything else. Our observations on the membrane lengths and the position in the uterus of the placenta in rupture of the marginal sinus suggest that many are almost marginal placenta previas.

It appears that a good deal of disentanglement of the syndromes of placenta previa, abruptio placentae, rupture of the marginal sinus, and "low implantation" remains to be done. A rigid standard of anatomic evidence for these diagnoses would result in a collection of cases of undoubted identity. Included in this diagnostic approach naturally would be palpation of the placenta during vaginal examination before delivery. In particular, the diagnosis of abruptio placentae has suffered from the assembling of cases in which the diagnosis was made because the total clinical picture suggested abruptio placentae, or that appeared to be the most likely diagnosis.

Summary and Conclusion

A separation of the syndromes of abruptio placentae and rupture of the marginal sinus of the placenta has been made by the establishment of strict anatomic criteria for these diagnoses. Justification of an anatomic basis for these diagnoses can be seen in the fact that 2 distinct clinical syndromes

emerged. The 2 syndromes were considerably different in symptomatology and risk to mother and fetus. There were 2 characteristics that abruptio placentae and rupture of the marginal sinus frequently had in common, namely, a tendency for the lesions to occur at the dependent portions of the placenta and a tendency to low implantation. This anatomic similarity suggests that under certain conditions one syndrome may be a precursor of the other.

References

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3. Nelson, R. L., and Brewer, T. H.: *Bull. Univ. Miami* 12: 87, 1959.

Discussion

DR. LOUIS M. HELLMAN, Brooklyn, N. Y.—In no obstetrical condition are comparable data more difficult to obtain than in premature separation of the placenta. This stems from the fact that specific diagnoses are largely clinical. Inasmuch as the perinatal mortality and maternal complications vary directly with the degree of separation, from total abruptio placentae with concealed hemorrhage to marginal sinus rupture, unless the various diagnoses are meticulously made, correlated statistics often differ widely and are virtually meaningless. Such a variation is well demonstrated in Tables I and II, which contain data on obstetric hemorrhage from 2 different localities. The Kings County Hospital statistics are roughly similar, both in prevalence of the constituent parts and in the perinatal mortality, to that presented by the essayist. On the other hand, the data obtained by Dr. Kohl from the Community Study in Hartford, Connecticut, show not a single instance of marginal sinus rupture and a very low perinatal mortality rate in abruptio placentae. One can only conclude that abruptio placentae is either quite mild in Hartford, or that little attempt was made to distinguish between the mild and the severe form. Even with the close agreement between the Kings County data and that from the Jackson Memorial Hospital, there seems to be a slight deficit in our diagnosis of rupture of the marginal sinus as compared with Dr. Ferguson's.

TABLE I. PREVALENCE OF ANTEPARTUM HEMORRHAGE (KINGS COUNTY HOSPITAL 1953-1957)

	PATIENTS		PERINATAL LOSS	
	NO.	%*	NO.	%*
Abruptio placentae	362	1.3	196	54.1
Other cause	353	1.3	86	24.4
Marginal sinus	123	0.5	21	17.1
Placenta previa	108	0.4	20	18.5
Ruptured uterus	34	0.1	7	20.6
Total	980	3.6	330	33.7

*Percentages are based on 27,502 deliveries.

TABLE II. PREVALENCE OF ANTEPARTUM HEMORRHAGE (COMMUNITY OBSTETRICAL STUDY,* 1959)

	PATIENTS		PERINATAL LOSS	
	NO.	%	NO.	%
Other cause	259	2.1	38	14.7
Abruptio placentae	257	2.0	59	23.0
Placenta previa	83	0.7	23	27.7
Ruptured uterus	4	0.2	0	0.0
Rupture of marginal sinus	0	0.0	0	0.0
Total	603	4.8	120	19.9

*Courtesy of Dr. Schuyler G. Kohl.

We, too, had one maternal death but it was in no way connected with the placental separation. We thus find ourselves in complete agreement with the philosophy of therapy expressed by the essayist, namely, that prompt delivery by cesarean section, if a short labor cannot be anticipated, has yielded excellent maternal results. The incidence of cesarean section in our series is 17.2 per cent. We have been liberal with our use of both transfusion and Pitocin, and we have not hesitated to perform cesarean section in face of shock, provided transfusion has already been started. The concepts upon which this program of therapy is based are prompt control of hemorrhage and a material shortening of the time from separation to delivery. Although, as Dr. Ferguson has stated, it is not proved that this lessens the risk to the mother, it seems to us quite likely that both the clotting deficit and the renal tubular damage are directly related to the length of time and the severity of the separation. This concept is supported by the relatively rare occurrence of hemorrhagic diathesis (15 in the author's series, 2 in ours) and the paucity of renal damage (3 patients in the author's paper and 3 in ours).

The possibility of additional infant salvage by increasing the incidence of cesarean section is interesting. In view of the high incidence of fetal death on admission and the prevalence of extreme degrees of prematurity, any marked decrease in perinatal mortality by this means seems unlikely. Furthermore, if I understand Dr. Ferguson's and my own philosophy of therapy correctly, cesarean section is directed first and foremost toward maternal and not fetal salvage.

When the Spanner concept of placental circulation was acceptable, the relatively high rate of fetal loss associated with rupture of the marginal sinus was explainable on the basis of interference with placental hemodynamics. Currently, Spanner is in disrepute and all investigations point to the correctness of Stieve's hypotheses that placental drainage takes place through veins located everywhere throughout the implantation site. This would seem to relegate the marginal sinus to an unimportant role in placental circulation and leave one hard pressed to explain the high fetal loss associated with such a minor degree of separation on any basis save prematurity. However, the loss of 2 of 70 term babies in the current series does not entirely permit this conclusion, for this is more than double the accepted rate. Perhaps during labor rupture of the marginal sinus, or any vein damage of the placental site, for that matter, allows the squeezing of blood from the intervillous space during uterine contractions. This could account for some fetal loss.

TABLE III. PREVALENCE OF PREMATUREITY IN TOTAL AND PARTIAL ABRUPTIO PLACENTAE AND RUPTURE OF THE MARGINAL SINUS (KINGS COUNTY HOSPITAL, 1951-1957)

	MATURE	PREMATURE	
		NO.	%
Total abruptio placentae	4	20	83.3
Partial abruptio placentae	127	257	66.9
Rupture of marginal sinus	72	81	52.9
Total*	203	358	63.8

$$\frac{*d}{s} = 3.30; p = .003 \text{ (total and partial abruptio placentae combined).}$$

TABLE IV. PREVALENCE OF PREMATUREITY IN ABRUPTIO PLACENTAE AND RUPTURE OF THE MARGINAL SINUS (JACKSON MEMORIAL HOSPITAL, 1955-1958)

	TERM	PREMATURE	
		NO.	%
Abuptio placentae	43	68	61.0
Rupture of marginal sinus	77	53	46.0
Total*	120	121	50.0

$$\frac{*d}{s} = 2.4.$$

Dr. Ferguson's very strict delineation between abruptio placentae and rupture of the marginal sinus on the basis of whether or not the clot grossly involves the decidual

plate cannot be confirmed on microscopic examination. In rupture of the marginal sinus there is always microscopic evidence of hemorrhage into the decidual plate and I believe that there is infiltration of this area with leukocytes and hemosiderin-laden macrophages. These facts would seem to indicate that the deficit is in this area and not solely confined to the marginal sinus.

The preponderance of available evidence points to a relationship anatomically, physiologically, and, to a certain degree, etiologically between complete and partial abruptio placentae and rupture of the marginal sinus—varying only in degrees of severity. Dr. Ferguson quite rightly calls attention to prematurity as one of the features of similarity between the two. If the contention of similarity with different degrees of severity is correct, however, the prematurity rate should show a decreasing incidence correlating with decreasing severity in the degree of separation. No only is this borne out statistically in our own data (Tables III and IV), but an identical trend occurs in Dr. Ferguson's data.

DR. RUDOLPH A. BARTHOLOMEW, Atlanta, Ga.—It would be unfortunate if the term "abruption" came to be applied to the occasional slight elevation of the placental margin by the clot formed at the site of, and due to, a rupture of the marginal sinus, thereby taking on some of the meaning inherent in the term "abruptio placentae." In the fresh placenta some degree of immediate differentiation is to be had by recognition of the soft consistency of the placenta nearest the point of sinus rupture versus the firmer consistency in case of true abruptio placentae at this site. An area of infarction always overlies an indenting clot, the latter being initiated by thromboplastin produced by the placental necrosis. Clear proof of this is obtained by gross and microscopic examination of the placenta fixed in formalin. The more massive the infarction, the greater the frequency of blood coagulation defects. I am confident the inclusion of this particular quest in the author's series would have shown a significant increase in the number of toxic cases.

It must be emphasized that the placenta, membranes, and adherent clot should be carefully laid aside until the delivery room duties are completed. The specimen is then examined. To send it to the laboratory at once may result in alteration of important relations through repeated handling.

When rupture of the marginal sinus occurs, the extravasating blood seeks the path of least resistance, which is outward beneath the more delicately attached membranes rather than inward beneath the margin of the placenta where greater resistance is encountered by reason of the fastening villi. One gets this impression firsthand in the relatively greater difficulty encountered in separating the more adherent placenta during cesarean section or manual removal of the placenta than in separating the more delicately adherent membranes.

In a previous analysis of bleeding during pregnancy it was found that one-third of all patients bled at some time during pregnancy. Two-thirds of those who bled did so during the first trimester. It is believed that the predilection to hemorrhage in the first trimester lies in the fact that the outer wall of the marginal sinus, very early in pregnancy, consists merely of decidua. Toward the end of the first trimester the expanding amniotic sac has filled the uterine cavity and contributed 2 supporting layers—the chorion and the amnion—to the outer wall of the sinus, plus the pressure of the amniotic fluid. Thereafter the frequency of hemorrhage is sharply reduced.

Nevertheless, rupture of the sinus continues to play a major role during the second trimester in the form of circumvallate placenta which represents the aftermath of retained blood from concealed or manifest bleeding in the first trimester.

In the third trimester during gradual formation of the lower uterine segment, the membranes in this region are subjected to increased stretch and tension produced by the upward pull of Braxton Hicks contractions and later descent of the presenting part. If the placenta happens to be situated low, as in any of the types of placenta previa (except the total), the marginal sinus is more apt to rupture and cause hemorrhage. This is particularly true in labor.

In an article which appeared in the *Southern Medical Journal* in November, 1958, Dr. Fish listed in Table IV the factors concerned in 347 consecutive cases of premature labor and late abortion. He noted that 49.5 per cent were due to premature rupture of the membranes and rupture of the marginal sinus (Table I). Actually the figure should be still higher since bleeding due to low implantation of the placenta and circumvallate formation should be included as examples of sinus rupture. By grouping these conditions, it is seen that 56 per cent of premature labors and late abortions are due to premature rupture of the membranes and rupture of the marginal sinus in equal proportion and are completely nonpreventable. Many of the remaining factors are also nonpreventable. Hence, a lowering of perinatal mortality will remain a challenge, but of doubtful attainment.

TABLE I. LATE ABORTIONS AND PREMATURE LABORS (347 CONSECUTIVE CASES)

	ABORTION	VIABLE	%
<i>Nonpreventable.—</i>			
Premature rupture of membrane	5	91	28
Rupture marginal sinus	21	55	28
Circumvallate placenta	5	4	
Placenta previa (except total)	3	9	
<i>Non- or Partially Preventable.—</i>			
Abruptio	2	7	3
Toxemia	0	12	3
Intrauterine death	21	18	11
Hydramnios	0	9	3
Twins	0	7	2
Assumed "term" cesarean	0	21	6
Unknown and miscellaneous	32	25	16
Total	89	258	100

Finally, I would like to express a plea that we cease designating the actively flowing blood in the wider portions of the marginal sinus as "lakes"—a term which implies stagnation. It is an appropriate term only if applied to the extravasation which may be formed as a result of rupture of the marginal sinus.

DR. FERGUSON (Closing).—Dr. Hellman remarked on the fact that in one hospital no cases of rupture of the marginal sinus were reported. I think that this is true of many hospitals. There are a number of reasons why this is so. The basic reason probably is that they are not aware of what the specimen looks like. Then, there is a tendency not to examine thoroughly the placenta of any woman who has had only a minor degree of bleeding.

Dr. Bartholomew raised the point about examination of the specimen that has been fixed in formalin. We have done that and I did not include it in this paper because we could not find any differences in the placentas that were prematurely separated and those that had a rupture of the marginal sinus. This may have been because we were not sure what to look for. Dr. Bartholomew has promised to come and visit us and point out the differences.

As to circumvallate placenta, I hesitated to comment on it because I have no control figures. It was found frequently in placentas that had rupture of the marginal sinus. I think that circumvallate placenta itself is more common than is credited. Until I can find someone in my department who will undertake this and obtain a control figure, my observations on circumvallate placenta and rupture of the marginal sinus, as combined lesions, will have to wait.

TROPHOBLAST IN THE CIRCULATING BLOOD DURING PREGNANCY*

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DEPORTATION of trophoblast, first described by Schmorl¹ in 1893, has been accepted in obstetrics as an example of the bizarre behavior of this tissue. Within its normal pattern of activity, trophoblast is capable not only of local invasion and mingling with maternal tissues, but of dispersal to distant sites as well, a feature which has often stimulated a comparison to the behavior of malignant neoplasms.

In his monograph, Schmorl attached significance to the fact that trophoblast was found regularly in the lungs of women dying of eclampsia (14 of 17 cases) but was not discovered in autopsy sections of 4 patients dying of other causes. In a later publication² this tendency was confirmed in a study of 158 deaths. Recently, Park³ reported similar findings in 53 of 120 cases. Bardawil and Toy⁴ noted trophoblastic embolism in 57 of 109 cases from the Boston Lying-in Hospital and described 2 instances of deportation of entire villi to the lungs.

These studies establish the fact that trophoblast does find its way to the pulmonary circulation of women who die during pregnancy, particularly in circumstances involving convulsions and intrauterine manipulation. However, lung sections from women dying of other causes show this tissue only occasionally. The problem of identification of trophoblast in this location is increased by the presence of megakaryocytes, large cells which bear a superficial resemblance to trophoblast. The findings in these tissues are seldom so widespread as to suggest that deportation of trophoblast is an ongoing, physiologic process in pregnancy. Instead, the possibility must be considered that terminal events in these cases are responsible for the occurrence of trophoblastic embolism.

The Placental Site

The histologic features of the placental site are of interest in regard to the possibility of trophoblastic deportation. In sections of placenta in the first trimester, one customarily finds very active proliferation of both cytotrophoblast and syncytium into the intervillous space, with only tenuous connections to the stromal base of villi. As pregnancy progresses, this pattern of

*Presented at the Eighty-second Annual Meeting of the American Gynecological Society, Hot Springs, Va., May 21-23, 1959.

outward growth diminishes, until at term the syncytiotrophoblast appears to be stretched rather thinly over the surface of the villi. Cytoplasm is scant, and masses of nuclei become prominent and are described as "knots." Nevertheless, even in the term placenta there are numerous small buds and sprouts of syncytiotrophoblast growing away from the surface of the villus. In tissue sections, these buds often appear to be floating free in the intervillous space (Fig 1).

The traditional view has been that these apparently detached masses of trophoblast maintain, *in vivo*, a tenuous connection with the surface of the villus, and that the avulsion of these fragments occurs from mechanical forces, as an occasional event in life, and more frequently from the handling incident to preparation of material for study. The proliferation of trophoblast is, of course, much more prominent in cases of hydatidiform mole. Thus, Hertig and Sheldon⁵ regarded "independent growth" as a significant feature in many of their collected cases, and, in a series of 206 cases which we have assembled for study, independent growth is a conspicuous finding in 96, or 47 per cent. Nevertheless, Hunt and Dockerty⁶ were not convinced that independent growth of trophoblast apart from villi occurs, and they noted that their findings could all be accounted for by tangential section and artifact.

The growth of trophoblast away from villous attachments and its migration into the blood stream as an active, normal pattern of behavior may occur in the

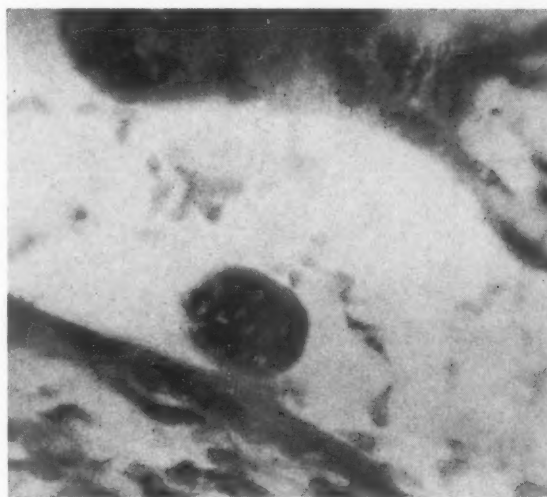


Fig. 1.—Tissue section from the placental site, term pregnancy. Syncytiotrophoblast sprouting from the surface of the underlying villus. ($\times 100$; reduced $\frac{1}{3}$.)

same manner as the known wandering of syncytiotrophoblast among the interstices of uterine muscle. If this is the case, then one immediate consequence of an active process of migration should be the appearance of trophoblast not only in the lungs but in the circulating blood during pregnancy.

The demonstration of cancer cells in the circulating blood by Engell⁷ and, more recently, by Moore and associates⁸ suggested the means by which trophoblast might be found in the blood stream. A preliminary report of this work has been made by Thomas and co-workers.⁹

Material and Methods

The method of making smears was generally similar to that employed by Moore. Ten milliliters of blood was drawn and immediately placed in a tube

containing 1.0 ml. of 1 per cent solution of ethylenediaminetetraacetic acid (Versene). The tube was rapidly inverted several times. Four milliliters of this sample was then placed in a centrifuge tube, and 1 ml. of 5 per cent fibrinogen added. Following incubation at 37° C. for 15 minutes, the supernate

Fig. 2.

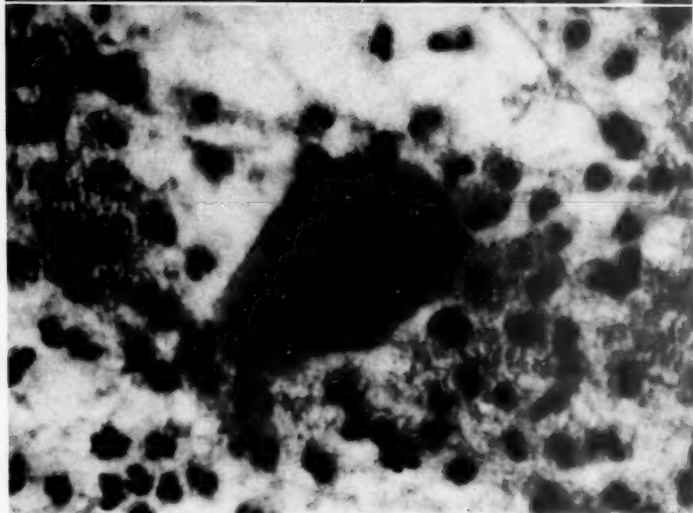
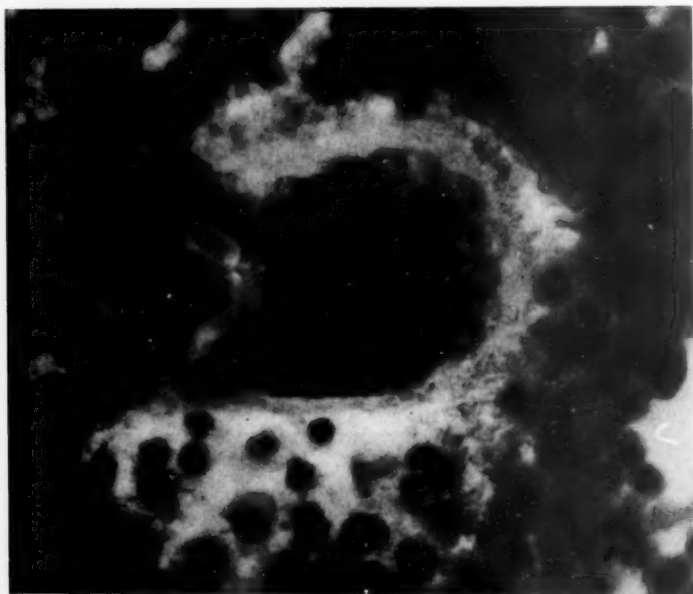


Fig. 3.

Fig. 2.—Large syncytial mass found in blood from a broad ligament vein, 18 weeks' gestation. (X475; reduced $\frac{1}{5}$.)

Fig. 3.—Syncytiotrophoblast found in blood from a broad ligament vein at the time of cesarean section, term gestation. (X475; reduced $\frac{1}{5}$.)

was drawn off and centrifuged for 5 minutes at low speed. The supernate was carefully drawn off and discarded and the remaining sediment smeared on 5 slides. These were fixed in equal parts of 95 per cent alcohol and ether and then stained by the Papanicolaou method.

Source of Blood Samples

1. *Placental Site.*—Samples taken from the placental site were taken at the time of cesarean section or laparotomy by passing a needle through the intact wall of the uterus prior to incision.

2. *Broad Ligament Veins.*—These samples were drawn from the large congeries of veins in the broad ligament, well away from the lateral border of the uterus at the time of cesarean section. In obtaining these samples, care was taken to avoid pressure and handling of the uterus.

3. *Samples From the Femoral Vein and Inferior Vena Cava.*—This approach was attempted in an effort to study the presence of trophoblast in the blood stream of patients not subjected to operation, at various stages of pregnancy, and on different occasions. Samples from the inferior vena cava were obtained through a polyethylene catheter threaded upward for a distance of 30 to 40 cm. at the time of a femoral venipuncture.

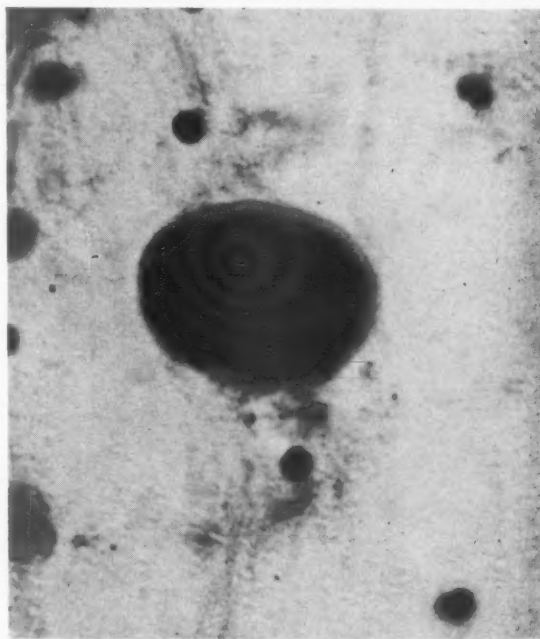


Fig. 4.—Cell from a broad ligament vein, term gestation. In the original smear, a fringed or "brush" cell border could be seen. ($\times 475$; reduced $\frac{1}{2}$.)

4. *Samples From the Ovarian Veins.*—These samples were obtained at the time of laparotomy for interruption of pregnancy and sterilization in 2 patients with advanced rheumatic heart disease.

5. *Peripheral Blood.*—Samples were drawn from the antecubital vein of 80 patients at varying stages of pregnancy, in the course of prenatal clinic visits.

6. *Controls.*—Each sample drawn from the inferior vena cava was accompanied by a sample from the femoral vein.* In addition, both femoral vein and inferior vena cava samples were obtained in nonpregnant individuals hospitalized for a variety of reasons, including several in whom the uterus had been removed.

Results

After the initial demonstration of syncytiotrophoblastic cells in the circulating blood, it was hoped that the variety of samples listed above would

provide information regarding the occurrence of this phenomenon in various stages of pregnancy, in the presence of various complications, as well as a rough approximation of the amount of tissue involved. A recent experience, however, has cast considerable doubt on the efficacy of the use of samples from the lower inferior vena cava as a means of finding these cells. Consequently, the results in this report will be largely descriptive, rather than statistical.

Blood drawn from the broad ligament veins has contained definite trophoblastic cells in 8 of the 13 cases studied thus far (Figs. 2-4). These cells are unmistakable, owing to their size and appearance, and are identical to syncytiotrophoblasts seen in tissue sections of the intact placenta, as well as to cells found in direct aspiration of the placental site. The latter technique has been employed in only a few instances, since it was felt that the direct trauma involved would cast doubt upon interpretation of results.

The typical syncytial trophoblastic cell as seen in the circulating blood is enormous, in the range of 100 to 200 μ , and is easily found. While exact counting of nuclei contained in these cells is difficult, estimates of 30 or more seem

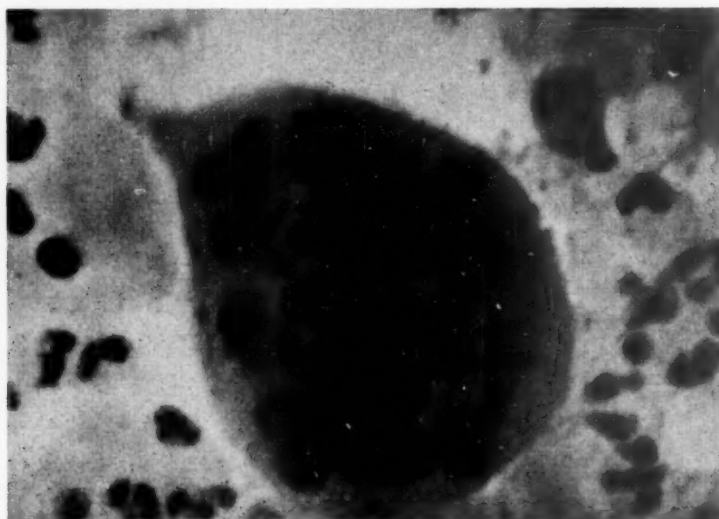


Fig. 5.—Syncytiotrophoblast, recovered from broad ligament vein, which retains the tear-drop shape frequently seen in syncytial buds at the placental site. ($\times 700$; reduced $\frac{1}{2}$.)

entirely reasonable. The cytoplasm is eosinophilic, and some individual cells are situated so that the typical brush border of syncytium is easily distinguished (Fig. 5). These cells have been found in a pregnancy of 18 weeks, in another case at 22 weeks, and at term in the remainder of positive instances.

Both cases in which samples were obtained in the second trimester showed much larger numbers of these cells in the blood than did the cases studied at term. Indeed, in the pregnancy terminated at 18 weeks, the frequency was such as to suggest the presence of at least one cell per milliliter of blood in the venous return from the uterus.

Blood drawn from the inferior vena cava of 33 patients at various stages of pregnancy including active labor has been studied. By painstaking search, we have discovered 3 clear-cut examples of syncytial trophoblast in samples of this kind (Figs. 6 and 7). At first we were inclined to attribute these rather disappointing results to the dilution of uterine venous blood by the circulatory return of the lower extremities, but recent experience has led us to question whether a major share of the uterine venous blood re-enters the general circulation at this level. In this regard, one case may be considered in detail:

Mrs. V. A., a 28-year-old gravida ii, para ii, was admitted in the twenty-second week of gestation, because of an exophytic carcinoma of the cervix, Stage I. Radical hysterectomy and pelvic lymphadenectomy were carried out. By this technique, we planned to ligate and

Fig. 6.

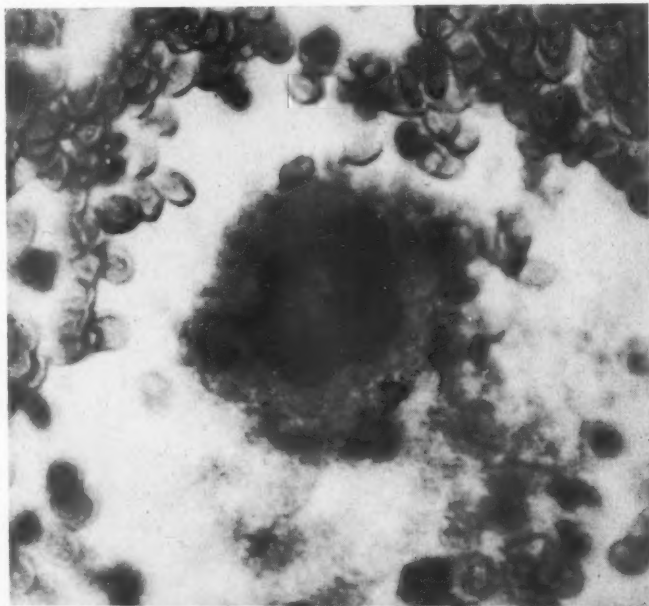


Fig. 7.

Fig. 6.—Trophoblastic cell recovered from the lower inferior vena cava. Pre-eclampsia, 38 weeks' gestation. ($\times 475$; reduced $\frac{1}{3}$.)

Fig. 7.—Term gestation, not in labor. This cell was recovered from the lower inferior vena cava. ($\times 475$; reduced $\frac{1}{3}$.)

sever the uterine vessels at their point of entry into the hypogastric artery and vein, close to the pelvic sidewall. To our surprise, the uterine vein was so small that its isolation proved a difficult task. The great engorgement of broad ligament veins was intimidating, and the ovarian veins were dilated to more than 1 cm. in diameter. The placenta

was implanted on the anterior wall of the body of this uterus. These findings suggested that the major route of return for uterine venous blood was by way of the ovarian vessels.

Samples of blood from both ovarian veins in this patient showed numbers of typical syncytial trophoblasts (Fig. 8), as did samples from the distended veins lateral to the uterus (Fig. 11). A sample from the inferior vena cava just above the pelvic brim, however, was entirely negative.

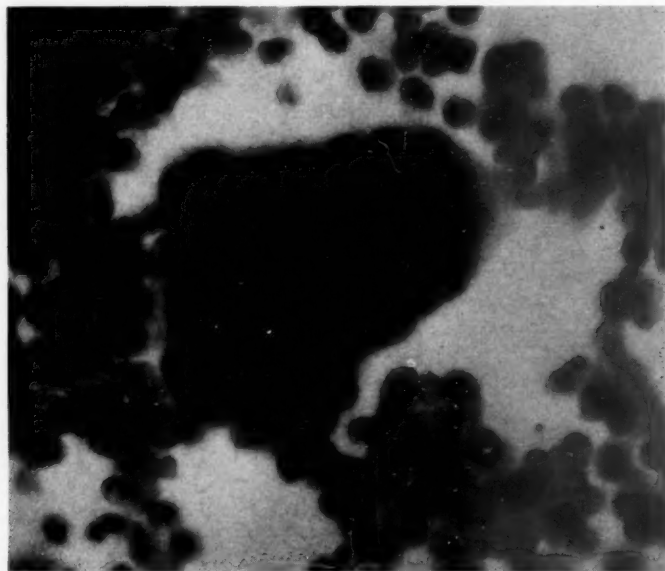


Fig. 8.—Large syncytial mass in ovarian vein sample. This patient had a radical hysterectomy for carcinoma of the cervix and was 22 weeks pregnant. ($\times 475$; reduced $\frac{1}{3}$.)

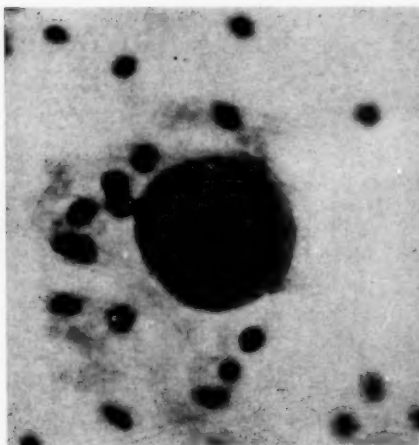


Fig. 9.

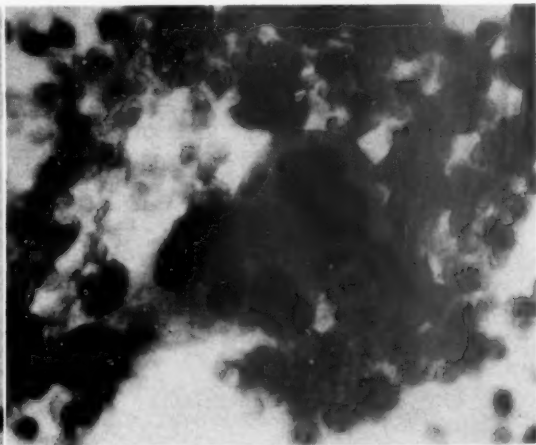


Fig. 10.

Fig. 9.—A typical megakaryocyte, found in blood from the inferior vena cava of a patient with pyelonephritis, 38 weeks' gestation. The nuclear mass is lobular, and the cytoplasm faintly basophilic.

Fig. 10.—Bone marrow smear from a nonpregnant individual. The cluster of megakaryocytes seen here closely resemble small, atypical trophoblastic cells.

Experience in this case has led us to question whether the major portion of uterine venous blood does return to the general circulation in the pelvis,

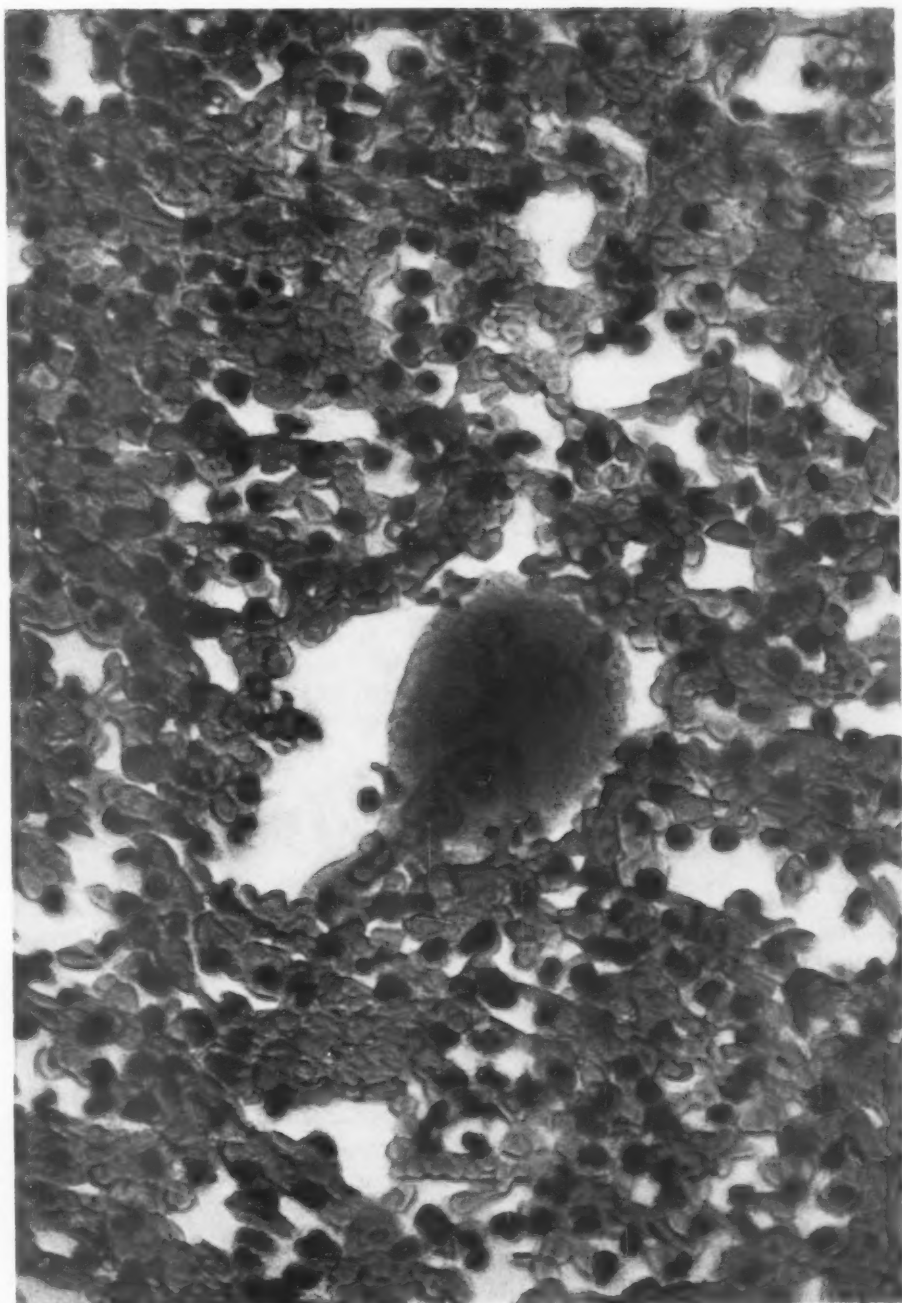
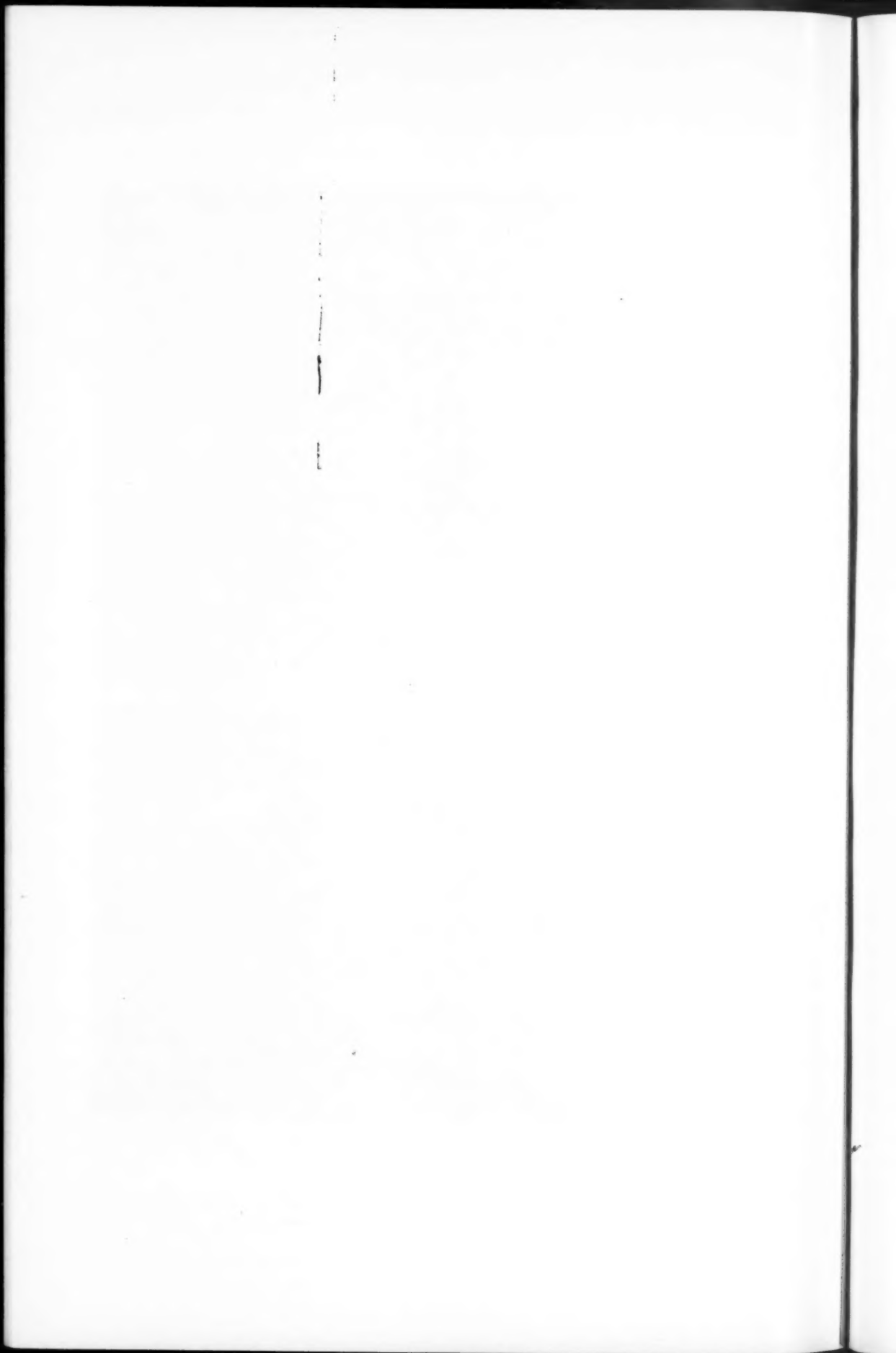


Fig. 11.—Trophoblast in blood from broad ligament veins, 26 weeks' gestation.



which was the area studied in our inferior vena cava samples, or at the higher level into which ovarian circulation is delivered.

Studies of the peripheral blood, drawn from the antecubital vein, have been uniformly negative in 80 pregnant women. No trophoblast was discovered in control samples of femoral vein blood, or in one instance in which blood was studied from the femoral artery.

Studies of control vena cava blood from 10 nonpregnant patients resulted in an incidental finding of some interest. In several patients, suffering from inflammatory conditions, numbers of large cells with lobed, pale nuclei were discovered (Fig. 9). These cells have been identified by comparison to smears of normal bone marrow as megakaryocytes, usually regarded as an inhabitant of the marrow and a source of blood platelets (Fig. 10). The presence of these cells in circulating blood has been infrequently reported in cases of leukemia and occasionally in cases of septicemia.

Megakaryocytes are found during pregnancy in samples of vena cava blood and in uterine venous blood as well. It is of interest to note that the most striking demonstration of megakaryocytes has been in a case of fulminating severe pre-eclampsia in the seventh month of pregnancy. A single slide, representing 1 c.c. of blood, contained more than 75 of these cells. While these cells bear no resemblance to the obvious syncytiotrophoblasts reported in this study, the possibility remains that megakaryocytes may be morphologically quite similar to smaller, atypical trophoblastic cells.

Comment

The demonstration of syncytial trophoblast in circulating blood drawn from a number of vessels draining the uterus, at various stages of gestation, makes it clear that the process of deportation may be regarded as a constant migration of this tissue into the blood stream during pregnancy. While our present methods do not permit an estimation in quantitative terms of the amount of tissue involved, nor of its relation to various complications of pregnancy, there is no doubt that this phenomenon is an accompaniment of normal pregnancy and not a result of death throes and intrauterine manipulation.

The migration of these cells into the blood stream may have profound significance as an explanation of the successful existence of a pregnancy. In a broad sense, pregnancy must be regarded as occupying the biologic role of a homograft, since both placental and fetal tissues have a genetic derivation different from the tissues of the maternal host. The genetic and immunologic requirements for successful homotransplantation of tissues are strict, and successful grafting between human individuals has generally been possible only in the case of monozygotic twins. It is clear that in pregnancy some fundamental means has been developed to escape the well-known fate of the homograft, so that existence of the conceptus for the period of gestation becomes immunologically possible.

Several obvious explanations for this apparent exception to homograft behavior may be considered. It may be that the fetal tissue which lies in juxtaposition to maternal tissues, the syncytiotrophoblast, is so embryonal and devoid of antigenic capacity that it is not recognized by the maternal host as an intruder. If one reviews the previllous ova described by Hertig and Rock,¹⁰ it is apparent that the rapid expansion of the freshly implanted ovum at the expense of maternal tissues occurs with virtually no morphologic sign of reaction to a foreign element. Yet the appearance of the placental site later in pregnancy suggests a sort of battleground, with the layer of Nitabuch and areas of necrosis representing a kind of no man's land.

Or it may be that the syncytiotrophoblast is recognized as foreign tissue by the mother, but the classic mechanisms of rejection are held in abeyance

by some intrinsic feature of the pregnancy which allows a sort of immunologic truce. Production or localization of cortisone or similar steroids by the placenta might provide a means for this mechanism of tolerance.

A third possibility is suggested by the work of Billingham, Brent, and Medawar,¹¹ in which tolerance to homografts was induced in animals by the prior injection of the donor's cells. It is conceivable that a similar mechanism is present in human pregnancy, and the migration of trophoblast into the blood stream throughout gestation is the means by which "tolerance" is achieved.

It is important to recall that sections of lung taken from autopsied cases of maternal death reveal embolic trophoblast only after careful search, and in many instances show no evidence of this tissue. This strongly suggests the existence of a mechanism for the destruction of these cells during their transit from uterus to lung, for otherwise the migration of trophoblast on the scale suggested by our findings would rapidly clog the pulmonary circulation and would produce symptoms at an early stage of pregnancy. Indeed, lung sections reveal no evidence of the arrival of this tissue in any such quantity, or of scars and reactive areas which would suggest the destruction of trophoblast in the lung as a continuous process in pregnancy.

A mechanism which may explain these findings is suggested by an experiment carried out by Thomas.¹² Fresh, teased villi were suspended in buffer, and a solution containing crystalline trypsin added. This produced lysis of the syncytiotrophoblast within minutes, leaving only a naked villous core. These results could be obtained with concentrations of trypsin as low as 1 gamma per cubic centimeter. Similar results were obtained by the use of chymotrypsin and human plasmin. An extreme susceptibility of syncytial trophoblast to the action of proteolytic enzymes in vitro is thus apparent, and if a similar mechanism exists in vivo the mysterious disappearance of this tissue before it reaches the lung is explained.

Thus, the demonstration of continual migration of syncytiotrophoblast, a tissue of fetal origin, into the maternal blood stream means that maternal tissues are exposed to those of the fetus and to fetal antigens, not only at the placental site but elsewhere as well. The implications of this phenomenon as a mechanism of tolerance are attractive, but they await studies of the intrinsic antigenic properties of trophoblast. This is the subject of further investigation at the present time.

Summary

1. Syncytiotrophoblast has been demonstrated in the circulating blood of pregnant women at various stages, from the eighteenth week of gestation to term.
2. The migration of trophoblast into the blood stream appears to be a normal process in pregnancy, apart from complications and circumstances leading to maternal death.
3. The biologic implications of pregnancy as a form of homograft are discussed, and the possible importance of trophoblastic migration into the blood stream as a means of achieving tolerance to fetal tissues is suggested.

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Discussion

DR. ROBERT A. ROSS, Chapel Hill, N. C.—The authors have presented an interesting study of a phenomenon which has been known to exist for many years. Using a method similar to Engell, Moore, and others, the authors secured blood from various veins and have documented their findings. Originally, and later, the presence of trophoblast in the lungs was thought to be associated with the toxemia of pregnancy or with a violent death. However, trophoblast has been found by us in fatal painless bleeding without labor or manipulation and in a woman at term who died quickly after being shot in the head while asleep. Certainly, one did not know that trophoblast was so frequently present in maternal circulation and in such numbers as suggested by the authors' findings.

It appears to this discussant that, as this fascinating study developed, the investigators became preoccupied with new facets and appealing possibilities, some of which are speculative, all attractive.

The word "migrate" suggests an active process which is somewhat speculative. The discrepancy between the finding of trophoblast in 7 of 12 patients' pelvic veins and in only 2 of 32 patients whose vena cava was entered might possibly be explained if blood were obtained above the ovarian and renal vessels. The position of the placenta in the uterus does influence return circulation in the pelvis. Data obtained during contractions of the uterus would be of interest and I am sure the authors will furnish these later. It is understandable that actual work on the primary project has prevented other than morphologic study to determine the viability of these cells. Attempts at culture, transplantation, and phase microscopy undoubtedly will be forthcoming. The sentence, "In a broad sense, a pregnancy must be regarded as occupying the biologic role of a homograft," opens myriad avenues of speculation and investigation: antigenic possibilities, tolerance, rejection, essential intrinsic steroid elaboration—all occur as possibilities. All are beyond the depth of this discussant. A mother's cell did participate in the conception, and all obstetricians have wish that the mother had rejected the placenta implanted on coils of intestines and other viscera in patients who have an abdominal pregnancy.

Studies of placental circulation by radiopaque material injection has demonstrated definite "spurts" of blood from small maternal arterioles into the intervillous spaces, and this action could effect the syncytiotrophoblast.

That trophoblast was demonstrated in greater number in the earlier pregnancies is hardly unexpected. The fundamental work of Hertig, Rock, and associates in the study of the developing trophoblast is properly recalled. One should remember that the placenta is "on the side" of the fetus, and this awesome cell is a fundamental unit of the functioning placenta. Under normal circumstances, the placenta is completely dedicated to the preservation of pregnancy. If altered by disease, trauma, or premature senility it can destroy fetus and mother.

The dramatic action of trypsin and chymotrypsin upon the villus as described by the authors makes one shudder at the possible effect one of the newer potent commercially prepared enzymes might have upon the placenta if administered to a pregnant woman who had a contusion, bruise, or swelling. It would be interesting to know if such a thing has been done.

The authors' work will aid in the study of the behavior of this normal cell, which most nearly approaches all the criteria inherent in a malignant cell.

DR. M. EDWARD DAVIS, Chicago, Ill.—Dr. Mohr from the Pathologic Institute of Düsseldorf, who is a research fellow in our institution, and I have been carrying out similar studies during the last year.

Fig. 1.

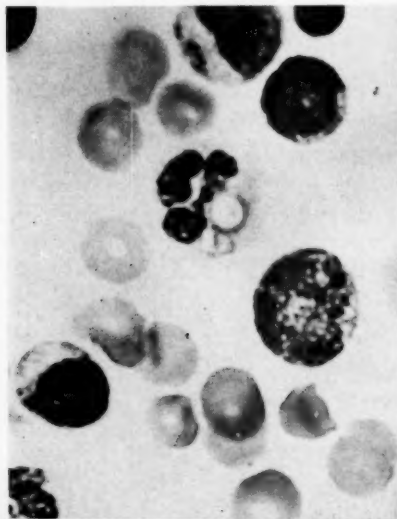


Fig. 2.

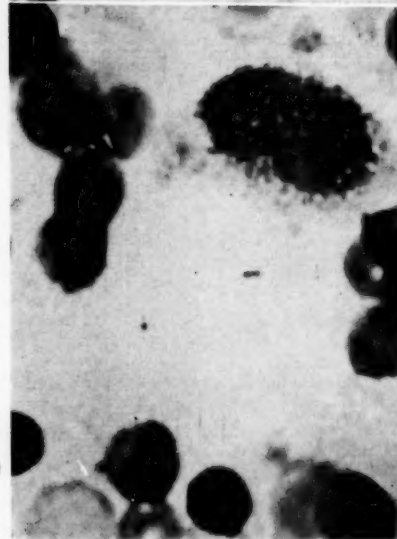
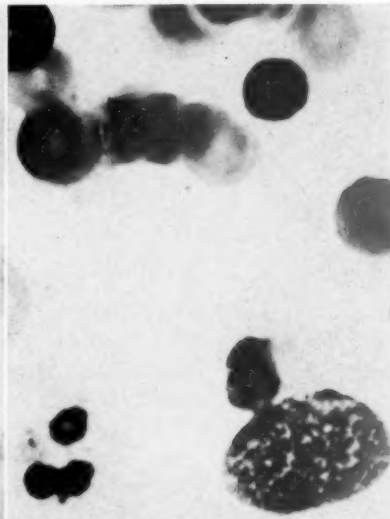


Fig. 3.

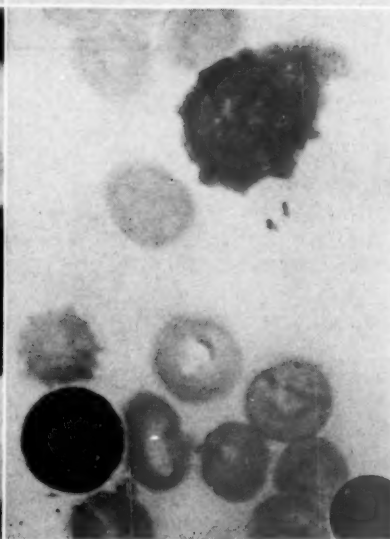


Fig. 4.

Fig. 1.—Smear made from a blood sample taken from a male and prepared by the Buckley technique in order to demonstrate the appearance of normal blood cells. ($\times 1,700$; reduced $\frac{1}{4}$.)

Fig. 2.—Smear made from a male blood sample into which chorionic cells scraped from a placenta were introduced. Note the large chorionic cell typical of those demonstrated in subsequent smears made from blood of pregnant women. The characteristics of syncytial cells found in maternal blood are quite different from the usual cell constituents of blood plasma. ($\times 1,700$; reduced $\frac{1}{4}$.)

Fig. 3.—Smear prepared from a blood sample taken from the broad ligament veins of a patient near term at cesarean section. The typical chorionic cell is surrounded by a rim of cytoplasm and is in a good state of preservation. ($\times 1,700$; reduced $\frac{1}{4}$.)

Fig. 4.—Smear from another specimen of blood taken at the time of cesarean hysterectomy. This chorionic cell shows considerable degenerative changes in comparison with the cell in Fig. 3. ($\times 1,700$; reduced $\frac{1}{4}$.)

Blood samples have been collected from women during pregnancy and at the time of cesarean section. Specimens of peripheral blood were obtained from the antecubital vein. Samples were drawn at the time of cesarean section from the grossly dilated veins of the broad ligaments, particularly prior to complete cesarean hysterectomy. Great care was exercised not to disturb the uterus or its contents and to avoid hematomas in the distended veins.

The isolation of the chorionic cells was accomplished by the Buckley technique. In this method the addition of fibrinogen to the blood sample results in the rapid sedimentation of erythrocytes by rouleaux formation. The sample is agitated and warmed by placing the tube under running water. It is then allowed to stand for about 20 minutes following which the supernatant fluid is removed and centrifuged at low speed for about 5 minutes. The supernate is then carefully drawn off and discarded and the sediment smeared on slides which were stained by Pappenheim's stain.

Our preparations have demonstrated single chorionic cells most often, and these are largely devoid of their cytoplasm. Dr. Douglas has been able to isolate rather large syncytial masses. Typical examples of the cells which we have isolated are illustrated in Figs. 1-4. The absence of cytoplasm surrounding the nuclei in many of the cells may be a manifestation of rapid autolysis by maternal immunologic mechanisms. However, it may have been brought about by our technique in the preparation of these slides. Dr. Douglas and his co-workers collected blood samples in ethylenediaminetetraacetic acid and incubated their specimens for 15 minutes. These two steps in their technique may preserve intact syncytial masses. We will explore our method of isolating chorionic tissue further. Suffice it to conclude at this time that the study of the transport of chorionic cells to the maternal circulation and its implications merit intense study.

DR. CARL P. HUBER, Indianapolis, Ind.—We in our department at the University of Indiana have been interested in a similar study, and in the last year have collected some 250 specimens, all obtained by antecubital venous puncture. Slides have been prepared similar to the method of Dr. Douglas and in approximately one third of these 250 specimens we have found large multinucleated cells. The blood specimens have been obtained from the sixteenth week of pregnancy through the immediate postpartum period. Most of them were obtained immediately following delivery of the placenta, and a considerable number 96 hours after delivery. We have found these cells in some of the specimens at all of these various times. The majority of the specimens contained these large cells. We are not completely convinced that we can be sure that some of these large multinucleated cells are not megakaryocytes or endothelial cells as well as perhaps trophoblastic cells.

DR. ERNEST W. PAGE, San Francisco, Calif.—Unlike Dr. Huber and Dr. Davis, I have no facts to offer nor any observations, but I was delighted to hear Dr. Douglas engage in a bit of speculation because this is a type of approach that has too long been unpopular. I will confine my remarks to the immunity mechanisms of which he spoke.

The one he favored was the Medawar phenomenon produced by an overwhelming amount of antigen, but this appears to be limited to the fetal stage and I know of no example in which such immunologic tolerance has been induced in the adult animal or in man. It seems to be limited to the prenatal or neonatal period before the development of the adult type of response to antigens. The second explanation, namely, the possibility of cortisone production by the placenta, is tenuous in that those who have investigated this problem seem to be in agreement that corticoids cannot be produced by placenta. It would seem unlikely, therefore, that these 2 mechanisms would account for preventing the maternal response to antigens.

The first concept that he mentioned, the absence of antigen from the syncytium, is supported by several studies; not only the absence of antigen but the syncytium may be capable of destroying any other antigen that enters its cytoplasm. This is borne out by the fact that antibodies transverse this cellular layer with ease but antigens are destroyed.

Therefore, the first of the explanations, namely, that if the syncytium picks up or contains antigen, it is destroyed by the cell itself, would appear to be the best explanation of why the host does not react to these cells by the production of antibodies or by rejection of the "homograft."

DR. LOUIS M. HELLMAN, Brooklyn, N. Y.—I would like to present one figure which supports the thesis raised by the essayist and also raises some questions (Fig. 1). This demonstrates the implantation site from a pregnancy between the sixteenth and the twentieth week. Below this implantation site is a maternal vein and growing in the vein is a thrombus of trophoblast cells. Not only do these cells partially block the vein but they have begun to replace the endothelium of the vessel itself. The free end of the thrombus is free in the vein. This side is from an early pregnancy and there is no difficulty in understanding the mechanism by which these trophoblast cells get into the circulation. Only a few would have to break off to supply the phenomenon which Dr. Douglas has described. So much for support of his thesis. The problem raised is this: if these cells are antigenic they

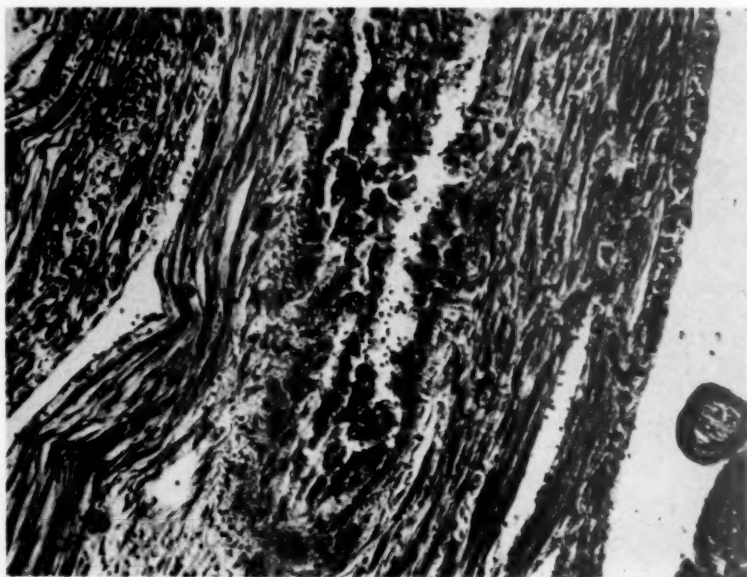


Fig. 1.

must be so at the site of the thrombus and should show some evidence of destruction. They appear to be perfectly healthy. Whether this is because they are actually at their site of growth or because they are not antigenic or because they possess the power of destroying antibodies which they lose once they are free in the circulation, I do not know. I would suggest, however, that they are not destroyed in the blood stream but in the specific organs to which they find their way, particularly in the lungs.

DR. DUNCAN E. REID, Boston, Mass.—Perhaps Dr. Douglas would comment further on the fascinating observations of Dr. Thomas. I was not certain whether the cytotrophoblastic as well as the syncytiotrophoblastic cells were destroyed when exposed to trypsin.

I would say that we must regard the mother as possessing the capacity to inactivate this migrating chorionic tissue. The failure to do so may explain the genesis of chorio-carcinoma, particularly those cases where the uterus at either hysterectomy or autopsy is found free of tumor. Here apparently the chorionic tissue remains dormant in some other part of the body sometimes for months or even years, and by some quirk of tissue-host relationship suddenly begins to grow with the resultant tumor rapidly reaching fatal

proportions. If plasmin could destroy or disturb the normal morphology of the cytotrophoblast, conceivably one might treat choriocarcinoma with some degree of success by activating plasminogen through the administration of streptokinase. Have you, Dr. Douglas, had any experience in this regard?

DR. DOUGLAS (Closing).—Dr. Ross, we were, of course, interested in searching for these cells in cases of toxemia. Blood drawn from the inferior vena cava of these patients does contain large, spectacular cells with multilobed nuclei, but on careful scrutiny these appear to be megakaryocytes, or possibly very atypical forms of syncytiotrophoblast. The unmistakable trophoblastic cell described in this paper was not found in the inferior vena cava blood of these patients. We have studied half a dozen patients during labor, and have found no difference in the specimens drawn during contractions, between contractions, or before the onset of labor.

Dr. Davis, I was intrigued by the appearance of the cells you described. They look as though disintegration is well under way. We have found similar cells, but only in blood drawn directly from the placental site. This suggests that the destruction of these cells is a rapid process indeed.

Dr. Huber's results in 250 cases are impressive. In our study of peripheral blood from 80 patients at various stages of pregnancy, we failed to find a single trophoblastic cell, but we will certainly review these smears again. It is possible that trophoblast could reach the peripheral circulation by way of vascular shunts, by-passing the pulmonary capillary bed. However, the trophoblastic cell is so large that we thought it would inevitably end up in the lung.

Dr. Page's remarks about the possibility of syncytium being capable of destroying antibodies are very provocative. This tissue is believed to harbor powerful proteolytic enzymes, but the mechanism of their activation or inhibition is, so far as I am aware, not known. It is conceivable that such an antibody is the means by which the invasive and destructive powers of trophoblast are held in check at the placental site, while on the other hand migration of trophoblast and its rapid disintegration in the blood stream are features of a strong proteolytic system directed to this tissue alone.

Dr. Hellman's comments regarding the presence of trophoblast lining vessels at the placental site are appropriate, since this is a likely source of some of the trophoblast in circulating blood and also because this represents direct contact of trophoblast with maternal endothelium, a tissue of great importance in the morphologic picture of homograft rejection. Dixon Boyd, of Cambridge, has described the peculiar appearance of these cells in detail. However, one does not encounter the picture of budding or sprouting growth of trophoblast in these vessels that is characteristic of this tissue as it separates from the surface of a villus. Perhaps further experience will allow us to relate the trophoblast found along the walls of these vessels with some of the atypical cells occasionally seen in blood smears.

Dr. Reid's comment regarding the possibility of setting in motion an active system of proteolysis by the use of streptokinase, is one that has occurred to us. Recently we tried this in a patient suffering from far-advanced choriocarcinoma, refractory to amethopterin. Although tests during administration of streptokinase showed the activation of a very potent system, the clinical course of the patient was unchanged and at autopsy the tumor areas did not appear to be affected.

FERTILITY AFTER CERVICAL DILATATION*

CARL T. JAVERT, M.D., NEW YORK, N. Y.

(From the Department of Obstetrics and Gynecology, College of Physicians and Surgeons, Columbia University, and the Woman's Hospital Division of St. Luke's Hospital)

THE literature on human sterility abounds with articles on the various infertility factors and how they may be overcome, while little is said about simple office dilatation of the cervix to promote fertility. The latest textbook on human infertility by Buxton and Southam⁵ does not even mention the method. The purpose of this article is to present my experience with 180 cases treated by cervical dilatation, including 158 fertile matings, of which 66, or 42 per cent, conceived; to review briefly the literature since 1930; and to speculate as to how dilatation works. I learned about the possibility of pregnancy following cervical dilatation from Dr. William H. Cary and it was at his suggestion that the present study was begun.

There are several articles in the medical literature which refer to conception after cervical dilatation or sounding of the uterus. Cary,⁷ van Tongeren,³⁷ Birnberg,¹ Siegler,³¹ Sharman,³⁰ and others have written on the subject. It would appear that conception takes place whether a cervical stenosis is present or not. The pregnancy occurs so promptly after the cervical dilatation that this simple method should receive most of the credit.

Technique of Dilatation

The cervical dilatation is usually performed on the first office visit as a part of the pelvic examination. The doctor-patient relationship begun in the office is continued in the examining room by asking further questions. A vaginal examination is performed to determine the size, shape, and position of the uterus and to ascertain the direction of the endocervico-uterine canal. The cervix is exposed with a sterile, medium-sized Graves speculum and Zephiran solution, 1:1,000 dilution, is applied to its surface. The smallest sterile Hanks' dilator, size 11 Fr., shown in Fig. 1 is then gently inserted into the external os and is pushed upward along the course of the endocervical canal as in Fig. 2. No local or topical anesthesia is administered although Siegler³¹ advocates the use of local infiltration procaine anesthesia. No tenaculum is placed on the anterior lip of the cervix. If the canal is easily traversed a second, next larger dilator is inserted in a similar manner. Frequently, the dilatation causes suprapubic pain which the patient says is similar to her menstrual cramps. This may be severe, especially in patients with severe dysmenorrhea, and such patients feel faint, become cold and clammy, and even become nauseated. The latter symptom may be due to reflex stimulation of the vagus nerve. These symptoms are transitory and the patient is quickly restored by inhalation of sweet spirits of nitre.

*Presented at the Eighty-second Annual Meeting of the American Gynecological Society, Hot Springs, Va., May 21-23, 1950.

Before the speculum is removed, a cotton tampon, dipped in Zephiran, is inserted into the vagina as a precautionary measure. There is often a slight amount of bleeding from the endocervix after the dilatation. The patient is instructed to remove the tampon that evening prior to intercourse. Coitus in knee-chest position is recommended. The vaginal speculum, dilator, and tampon are always shown to the patient before they are inserted into the vagina.

The scattergram shown in Fig. 3 reveals the various days of the menstrual cycle on which the dilatations were performed that were followed by conceptions. In some cases dilatation was done as early as the sixth day and in others as late as the twenty-fifth day. There were 8 dilatations late in the cycle, and 3 of these were in patients who may have been pregnant at the time of the dilatation, yet they were not harmed by this procedure. The remaining 5 cases were in women with unusually long cycles and probably should not have been included in the scattergram.

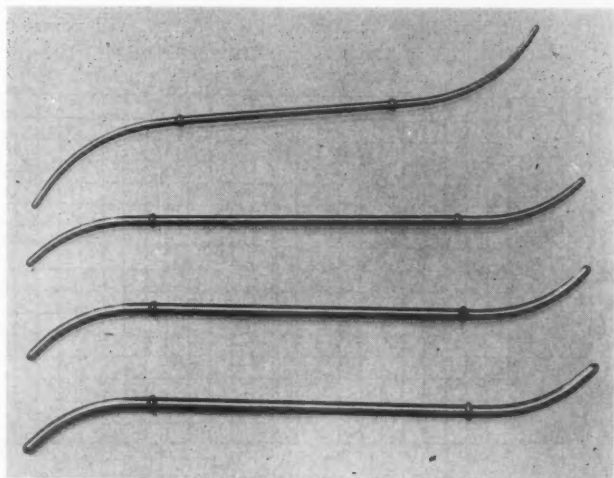


Fig. 1.—A set of Hanks' cervical dilators ranging from 11 Fr. upward.

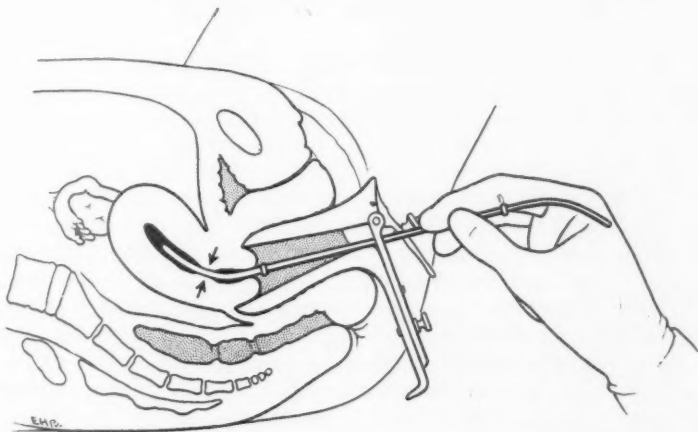


Fig. 2.—Technique of office dilatation of the cervix.

When pregnancy did not occur, the dilatation was repeated in 2 or 3 months. It was scheduled and performed as soon as possible after completion of the menstrual period.

There were no control cases in the ordinary sense of the word, unless one wishes to so consider a group of 18 patients in whom a pelvic examination only was performed, as shown in Table I. These patients did not conceive. Sharman³⁰ found that 10 per cent of his patients conceived after a bimanual pelvic examination.

Materials and Methods

This study is based on 221 private patients who had the complaint of infertility for one year or more. Most of them were seen during the past decade and up to April 1, 1959. Further cases will be added to this study, which is still in progress. A summary of the types of treatment administered either in the office or in the hospital is shown in Table I. Less than one-fourth of the patients were also treated in the hospital. Most had office dilatation of the cervix performed one or more times. There were 149 cases of dilatation in the office as well as an additional 31 following various surgical procedures in

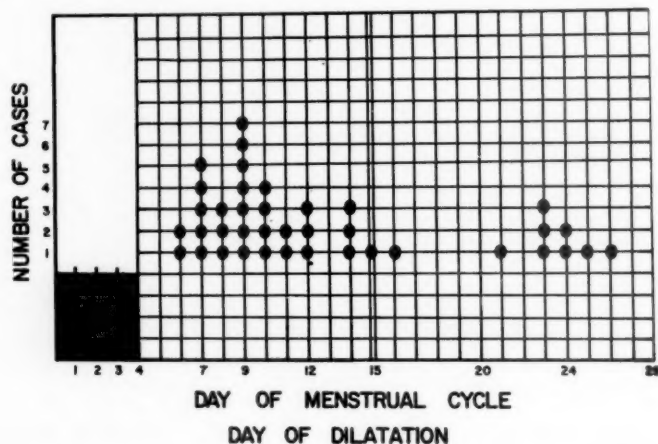


Fig. 3.—Scattergram showing day of menstrual cycle that 39 cervical dilatations were done which were followed by conception within one month.

the hospital, for a total of 180 dilated cases. Office dilatation proved to be the chief method of treatment since many of these patients had been subjected to insufflations and hystero-grams prior to the present investigation. Consequently, this is selected material in that it does not consist only of women seeking an opinion for the first time. About half of them had seen other physicians and about half were new patients being seen for the first time. The latter provided most of the tubal and sperm factors revealed by the investigation. However, 28 must be considered "refractory" according to the definition given below. The vast majority of patients, namely 158, represented fertile matings. One or more cervical dilatations had been performed on them and they must be regarded as potential candidates for pregnancy. Many of them had been told previously by other physicians that there was no obvious reason for their infertility. Naturally, one might expect good results with the simplest treatment in such a group and this proved to be the case.

Table II lists the various infertility factors found in the 221 patients, according to whether they had primary (never conceived) or secondary (infertile after previous pregnancy) cases of sterility. Nearly two-thirds of the

cases were of the primary variety. Table II shows the dominating infertility factor in each case. To be sure, most of the cases had 2 or more of these factors, i.e., cervical stenosis and a psychologic factor or stenosis and scanty periods. Under such circumstances, my opinion prevailed as to which single, anatomical factor was most important. The main purpose of this tabulation was to determine the incidence of cervical stenosis in this material. It was found to be the most frequent defect, and yet it was present in only 35 cases, or 16 per cent. This is of major significance when one considers that office dilatation was performed in many more, namely 180 or 81.5 per cent; obviously on many who did not have stenosis. This opens up many possible avenues for "modi operandi," which will be discussed below. The 4 leading infertility factors in the total material were as follows: cervical, refractory, uterine, and spermatie, in that order.

TABLE I. TOTAL NUMBER OF CONCEPTIONS IN ENTIRE SERIES OF 221 PATIENTS ACCORDING TO TYPE OF TREATMENT RENDERED

TYPE OF TREATMENT	TOTAL NO.	OFFICE CERVICAL DILATATION IN ADDITION*	TOTAL CONCEPTIONS	
			NO.	%
<i>Office Procedures.—</i>				
Pelvic examination and cervical dilatation	127	127	53	
Pelvic examination and pessary	19	16	4	
Pelvic examination only	18	—	0	
Pelvic examination and erosion coagulation	5	3	3	
Pelvic examination and treatment for vaginitis	3	2	0	
Pelvic examination and polypectomy	1	1	0	
Total	173	149	60	34.6
<i>Hospital Procedures.—</i>				
Curettage, tubal in- sufflation	36	24	9	
Myomectomy	5	3	3	
Tuboplasty	2	1	0	
Emmet-Lash operation	2	1	1	
Lash operation alone	2	2	1	
Uterine suspension	1	0	0	
Total	48	31	14	29.2
Grand total	221	180*	74*	33.4

*Sixty-six conceptions followed cervical dilatation as the only or most recent treatment administered.

The dilatation was done in 20 patients soon after intercourse in order to give the sperm a "push." Fraenkel¹⁰ has advocated the use of tubal insufflation for a similar purpose. This technique did not appear to have any special value in these cases.

The amount of pain produced by the dilatation was measured by the Hardy-Wolf-Goodell dolorimeter in 10 patients. They sustained an average pain intensity ranging from 2 to 6 dols. The former is like a menstrual cramp and the latter may be compared to a moderately severe labor pain. This pain is, of course, transient. Most of the patients did not mind it at all, others did not like it too much but remained stoic, while a few cried out and did not like it at all. The latter group had other mannerisms, as discussed below under "psychosomatic aspects," which seemed to signify that they were not really interested in motherhood.

TABLE II. DISTRIBUTION OF 221 CASES AS TO PRIMARY AND SECONDARY STERILITY AND THE PREDOMINANT INFERTILITY FACTOR IN EACH CASE

INFERTILITY FACTOR	TYPE OF STERILITY		TOTAL	
	PRIMARY	SECONDARY	NO.	%
Cervical stenosis	25	10	35	15.8
Uterine factor	19	10	29	13.1
Endometrial factor	4	2	6	
Tubal factor	4	5	9	
Ovarian factor	1	0	1	
Peritoneal factor	2	1	3	
Vaginal factor	2	1	3	
Coital factor	1	1	2	
Sperm factor	11	4	15	6.8
Husband apathetic	2	0	2	
Psychological factor	0	3	3	
Oligomenorrhea	7	4	11	
Hypothyroidism	2	1	3	
Pregnancy failure factors*	0	15	15	6.8
Age factor	4	2	6	
Orthodox factor	2	0	2	
Refractory (complete work-up)	24	9	33	14.9
Undifferentiated (incomplete work-up)	28	15	43	
Total	138 (62.4%)	83 (37.6%)	221	

*Stillbirths, abortions, anomalies.

Results

Table I reveals that pregnancies occurred in 74 of the entire series of 221 infertility patients, or 33.4 per cent, which is a respectable percentage. The results were slightly better in office patients than in hospitalized patients for the obvious reason that the latter had more serious problems. These were not necessarily relieved by the hospital procedures, for 31 out of the 48 patients hospitalized also had dilatation subsequently in the office. Only 12 conceptions followed the hospital procedures per se. However, there were 66 conceptions following office cervical dilatation as the primary or most recently administered procedure in 180 cases or 36.7 per cent. This may be corrected because of sperm deficiency in 15 cases and closed tubes in 7 for a final total percentage of 41.8 per cent as shown in Table III.

TABLE III. DISTRIBUTION OF 180 CASES OF CERVICAL DILATATION AS TO CONCEPTIONS AND TYPE OF STERILITY

TYPE OF STERILITY	NO. OF CASES	CONCEPTIONS	
		NO.	%
Primary	117	42	35.9
Secondary	63	24	38.1
Total	180	66	36.7
Corrected for tubal and sperm factors	158	66	41.8

The relationship of the time of the dilatation to the time of conception is shown in Table IV. There were 39 conceptions within one month of the cervical dilatation, or 59 per cent. A scatter of these dilatations according to the day of the menstrual cycle is shown in Fig. 3.

Table V gives the number of dilatations employed in the total number of 180 patients; most of them had only one dilatation and 78.8 per cent conceived.

However, perseverance had its reward, for 14 additional pregnancies followed 2 or more dilatations. Of the patients whose cervixes were dilated, 114 did not conceive. Further investigation revealed some of the infertility factors listed in Table I, including closed tubes and a sperm factor. A fair number found the dilatation too painful, and in a few cases seen recently, sufficient time has not elapsed to appraise results.

TABLE IV. CONCEPTIONS IN RELATION TO TIME OF OFFICE DILATATION

CONCEPTIONS	TIME AFTER CERVICAL DILATATION (MONTHS)
39 (59.1%)	1
8	2
5	3
1	4
4	5
5	6
4	Over 6

TABLE V. CONCEPTIONS ACCORDING TO THE NUMBER OF DILATATIONS PER PATIENT

NO. OF DILATATIONS	TOTAL NO. OF CASES	CONCEPTIONS	
		NO.	%
1	143	52	78.8
2	31	10	15.2
3 or more	6	4	6.0
Total	180	66	100.0
Corrected for fertile matings	158	66	41.8

Follow-up studies of 53 patients have revealed 24 additional pregnancies; 16 occurred without further medical aid, but 8 followed another cervical dilatation. These 8 conceptions bespeak the value of cervical dilatation in patients with secondary sterility. Three patients required an Emmet's repair of cervical lacerations before they became pregnant again.

Table III reveals a slightly higher percentage of conception in patients with secondary sterility than in those with primary sterility. The difference is slight, yet it may signify that cervical stenosis is not the factor that is overcome by the dilatation as one may suppose. There are probably other avenues to be considered, which will be done below under "Modi Operandi."

A considerable number of the sterility patients who did not conceive volunteered the information on subsequent office visits that their dysmenorrhea had been relieved. Some have returned for periodic dilatations even though the husband has azospermia. Several infertility patients with irregular menstrual cycles had a period on the tenth to the twelfth day following the office dilatation. Many conceived within a month and their days of dilatation are shown in Fig. 3.

Spontaneous Abortion and Other Types of Delivery

There were 74 pregnancies in the entire series of 221 patients or 33.4 per cent, as indicated in Table I. Of this number, 14, or 19 per cent, ended in spontaneous abortion, as shown in Table VI. Other authors have reported an increased incidence of miscarriage in the infertility patients. The usual incidence is about 10 per cent. For the past 5 years, I have employed the habitual

abortion regimen in sterility patients hoping to keep the number of spontaneous abortions at a minimum. The number of cases so treated is too small for any conclusions, but this approach may have merit.

There were 44 full-term deliveries, of which 25, or 58 per cent, were spontaneous. A problem encountered in labor with some frequency in the patients with a cervical factor was cervical stenosis. Pitocin and Releasin were helpful in some, while cesarean section was resorted to in a few others because of cervical dystocia. The data pertaining to types of delivery can be found in Table VI.

TABLE VI. TYPE OF DELIVERY IN 74 PREGNANCIES IN THE ENTIRE SERIES OF 221 STERILITY PATIENTS

Spontaneous abortion	14	(18.9%)
Premature delivery	2	
Full-term delivery	43	
Spontaneous	25	
Low forceps	7	
Midforceps	4	
Cesarean	7	
Ectopic pregnancy	1	
Undelivered	9	
Term delivery elsewhere, type unknown	5	
Total	74	
Fetal mortality	0	

Refractory Sterility Patients

For the purpose of this study, a refractory sterility patient has been defined as one who has not conceived for one year or longer after trying to do so prior to the first investigation. She is 40 years of age or younger and has a fertile and potent husband. She has no demonstrable physical or gynecological defect and has had a complete infertility work-up. She has been seen by several competent doctors and has been followed by them for an additional year or more, so that the total period of infertility has been 2 years or more.

TABLE VII. "REFRACTORY" STERILITY PATIENTS

Total No. of cases	221	
No. considered "refractory"	33	(15%)
No. having office dilatation of cervix	28	
No. conceiving thereafter	8	(28%)

Out of the entire series of 221 patients, 33, or 15 per cent, were considered "refractory" by the above standards (Table VII). Of these, 28 underwent cervical dilatation as part of their initial office work-up and 8 or 28 per cent conceived. Some of these patients had been investigated by several outstanding sterility experts.

Comment

There are several possibilities to be considered as to how mechanical dilatation of the cervix brings about a pregnancy. The *modi operandi* may be listed as follows:

Modi Operandi.—(1) Stimulation of ovulation, (2) dilatation of a stenotic internal os, (3) improvement of the cervical mucus, (4) facilitation of sperm migration, (5) enlargement of the infantile uterus, (6) psychosomatic aspects, (7) coincidence.

Stimulation of Ovulation.—The literature mentions various methods of inducing ovulation in the human female. These include: administration of gonadotrophic hormone, diathermy, irradiation of the pituitary and ovaries, relief of psychic tension, and mechanical dilatation of the cervix. Only the last-named procedure was employed in this study. No direct evidence of ovulation was obtained in these patients. No endometrial biopsies were performed and only a few patients kept temperature charts. Fig. 4 shows the chart of one patient who did keep a good record.

Ovulation can be diagnosed positively only by actual observation of the ovaries either by culdoscopy or by laparotomy. Neither of these methods was too feasible in these patients although this phase of the study needs further consideration. The effect of dilatation of the cervix on varying days of the menstrual cycle was studied in 24 other patients for whom a laparotomy had been planned anyway. Enlarged, hemorrhagic follicles were found in the ovaries of 4 patients; a biopsy was taken of one of these follicles and it was thought that the dilatation coincided with a spontaneous ovulation.

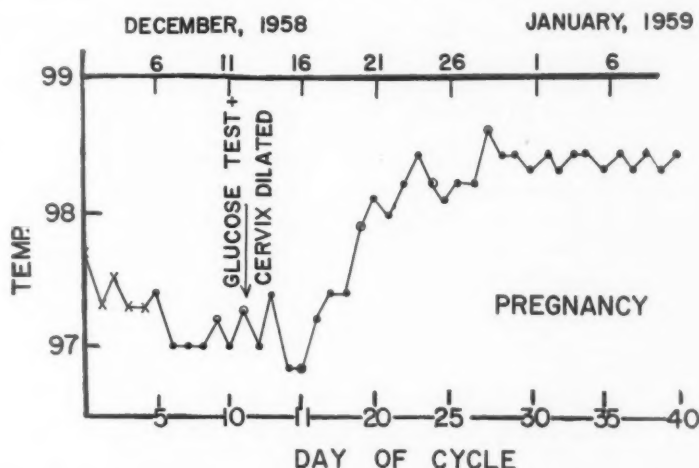


Fig. 4.—Temperature chart of a patient with positive glucose test and dilatation of cervix on eleventh day of menstrual cycle, followed by pregnancy.

Parkes²⁶ referred to mechanical ovulation in 1929 and postulated that the ovary was stimulated by the cervical dilatation through the anterior lobe of one pituitary. Soon thereafter, a number of reports appeared on sounding the uterus or dilating the cervix. Some of these have been collected in Table VIII and of 705 patients, 229, or 32.5 per cent, conceived. Cary⁷ was first with a series of 100 sterility cases in 1933 containing 5 patients who had had dilatations, 4 of whom became pregnant. Stein and Leventhal³³ had published an article previously that did not mention the procedure. Birnberg¹ reported 30 cases of dilatation in 1937, in which 3 pregnancies occurred. The same year, van Tongeren³⁷ reported sounding of the uterus in 185 patients; 88, or 48 per cent, conceived. Birnberg¹ detected a positive prolan response in the urine within 30 hours after the dilatation in many of his cases. He said in a recent communication² that many physicians still believe that this is a good way to stimulate ovulation. Siegler³¹ dilated the cervixes of 26 women up to 1944 and obtained 13 pregnancies. He dilated the cervixes of 5 other women with normal cervixes, and 3 pregnancies followed. These reports in the literature were followed by Sharman's study³⁰ in 1952 of various therapeutic experiments in

infertility. He sounded the uteri of 253 patients of whom 45, or 18 per cent, became pregnant. Gray¹³ reported a small series of cases in 1954. My current success rate is 41.8 per cent, as shown in Table III.

TABLE VIII. COLLECTED CASES OF PREGNANCY AFTER SOUNDING THE UTERUS OR DILATION OF CERVIX

AUTHOR	YEAR	NO. OF DILATIONS	PREGNANCIES	
			NO.	%
Cary ⁷	1933	5	4	80.0
van Tongeren ^{37*}	1937	185	88	47.6
Birnberg ¹	1937	30	3	10.0
Siegler ³¹	1944	31	16	51.6
Sharman ^{30*}	1952	253	45	17.7
Gray ¹³	1954	21	7	33.0
Javert	1959	180	66	36.7
Total		705	229	32.5

*Sounding of uterus.

Kurzrock²² commented in 1953 that cervical dilatation may lead to follicle stimulation and ovulation resulting in a pregnancy, which caused Weisman⁴¹ to comment: "Such a procedure has been long known to evoke ovulation in a rabbit. The occurrence in human beings is not generally established."

TABLE IX. COLLECTED CASES OF PREGNANCY AFTER TUBAL INSUFFLATION

AUTHOR AND YEAR	TOTAL NO. CASES	PREGNANCIES	
		NO.	%
Rubin ²⁹ (1945)	2,261	590	26.1
King and Herring ¹⁹ (1949)	82	28	34.1
Buxton and Southam ⁵ (1958)	859	99	11.5
Total	3,202	717	22.0
Buxton and Southam ⁵ (1958) (without tube test)	422	82	19.7

TABLE X. TIME OF CONCEPTION IN RELATION TO TUBAL INSUFFLATION IN RUBIN'S CASES²⁹

PREGNANCIES	TIME AFTER INSUFFLATION (MONTHS)
158 (41.8%)	1
70	2
37	3
38	4
35	5
40	6

TABLE XI. TIME OF CONCEPTION AFTER SOUNDING OF THE UTERUS IN 185 CASES FROM VAN TONGEREN³⁷

PREGNANCIES	TIME AFTER SOUNDING OF UTERUS (MONTHS)
21 (23.9%)	1
21	1-2
19	3-4
12	5-6
15	Over 6

Dilatation of a Stenotic Internal Os.—Table IX shows data from several articles in the literature, on several thousand women after tubal insufflation. There were 717 pregnancies in 3,202 patients, or 22 per cent. Rubin²⁹ had performed insufflation in most of these cases and obtained 590 pregnancies.

Most of them occurred within one month after the insufflation, as indicated in Table X, a fact not emphasized by Rubin.²⁹ Perhaps his success, which is ordinarily attributed to "opening the tubes," should be credited to opening the cervix with the insufflation cannula. Yet, van Tongeren,³⁷ by sounding the uterus, produced pregnancies in only 24 per cent of his patients within a month of this procedure (Table XI). King and Herring¹⁹ did not employ dilatation per se, but they reported 28 pregnancies in 82 patients following tubal insufflation or hysterosalpingography. Buxton and Southam⁵ noted almost as many pregnancies with the tube test as without it in their cases, as revealed in Table IX. Turner and associates³⁶ observed 60 pregnancies after cervical dilatation and curettage, or 38.8 per cent. Van Tongeren³⁷ attributed his success to dilatation of the internal and external ora. Reynolds²⁷ has observed pregnancies after simple passage of the uterine sound. Cary⁸ attributes most of the pregnancies obtained under such circumstances to the coincidental cervical dilatation.

According to Zondek,⁴³ cervical stenosis and sterility are related. Grant¹² says the "cervical factor" is the commonest one. Overstreet²⁵ claimed an incidence of 70 per cent in his cases, surpassing the tubal, uterine, and male factors. Veharskari³⁹ determined a cervical factor in only 2 per cent of his cases. My incidence of cervical stenosis, shown in Table II, was 16 per cent.

I was impressed with stenosis of the internal cervical os as a sterility factor in 35 patients, of whom 31 conceived after dilatation. It is seldom mentioned as such, while most articles mention a tight external os as "cervical stenosis." Among my cases of dilatation, varying degrees of stenosis of the internal os were encountered. Many of these patients had been told by other physicians that there was nothing wrong. I have relieved infertility by dilating the internal os, and I have seen it produced by the Lash operation on habitual abortion patients in order to tighten an incompetent internal os. Four such patients developed secondary sterility and severe dysmenorrhea after this operation. After their iatrogenically produced stenoses of the internal os were treated, 3 patients became pregnant.

Improvement of the Cervical Mucus.—The importance of a clean, watery cervical mucus has been stressed by most present-day writers on infertility. I have observed a relative scarcity of endocervical mucus in some of the infertility patients, especially in those with cervical stenosis. Cary⁸ frequently stressed the value of repeated dilatations in stimulating the flow of mucus. This effect has been recently mentioned also by McLane, Gepfert and Sweeney.²³ I wish to emphasize the importance of blood in the canal after dilatation as providing a medium for sperm migration when mucus is deficient.

Facilitation of Sperm Migration.—One of the curative methods for the relief of sterility is the dilatation of atresic or stenotic cervixes to facilitate sperm migration. The use of the stem pessary for this purpose has been advocated by many, including Frost¹¹ and Cary.⁸ However, neither cervical dilatation nor the stem pessary were mentioned by Buxton and Southam⁵ in their recent book on infertility. While Childs⁹ said in his book (1922) that spermatozoa were microscopic and therefore a small or minute opening in the cervix did not cause sterility, he nevertheless advocated the correction of cervical stenosis. Siegler³¹ mentioned 410 cases in his book, of which 157 had cervical factors including stenosis. This condition, as well as ample mucus, is an important factor in sperm migration.

Enlargement of the Infantile Uterus.—Vignes⁴⁰ advocated dilatation of the cervix in order to increase the size of an infantile uterus. Others employ hormone therapy for this purpose. Many women with small uteri have no difficulty in conceiving. This theory can probably be ignored in terms of "how cervical dilatation works."

Psychosomatic Aspects.—Psychogenic sterility is receiving attention today, along with the time-honored cervical, tubal, uterine, and endocrine factors, as a cause of sterility. It has been discussed by Kamman,¹⁸ Kroger and Freed,²¹ Siegler,³¹ and Buxton and Southam.⁵ Bos and Cleghorn³ say that the diagnosis is usually made by exclusion. I believe that the cervical dilator helps to detect some of the cases by evoking a certain reaction pattern in the patient. As mentioned above under Materials and Methods, cervical dilatation evokes 2 to 6 dols of pain according to Javert and Hardy.¹⁶ Most of the 180 women who had dilatations performed left the examination room with a smile. A few said "never again." Some had a better reaction pattern than others. A few developed cervical dystocia in labor. The relation of pelvic organ innervation and the various neuroendocrine mechanisms have been described by Harris¹⁵ and also by Javert.¹⁷ Other evidence of a hidden antipathy to fertility is offered by those patients who insert absorbant tissues into the vagina immediately after coitus or douche immediately after coitus. These practices are commonly indulged in by sterile and infertile women. Not every woman complaining of sterility actually wants a child. It has been estimated that one infertility patient in 5 does not follow the doctor's advice.

The Hanks' dilator is as valuable as a lie detector in the study of the sterility patient. It helps to disclose their psychoneurotic attitudes long before the onset of pregnancy and labor. It helps to detect those women who do not want to conceive, yet go through all sorts of tests and operations. Some prefer the "pleasure of a miscarriage." They merely wanted to show the world that they could become pregnant. Others behave very badly in labor and utter "never again."

Sometimes the sterile woman is satisfied with her lot. She revels in the extra attention and pity that such a state evokes from her friends. For what reason does such a patient come to the physician? Pressure from husbands, relatives, friends, and in-laws drive some of them to the doctor. Yet they do not always follow his advice. Some hope to learn that their marriage is sterile so that they can stop using contraception. Others are trying to avoid the social stigma of sterility. About 20 per cent of the patients are said to belong in the category of psychogenic sterility.

Coincidence.—Pregnancy occurred within a month after cervical dilatation in 59 per cent of the dilated cases. This is almost twice as good as the success rate of 30 per cent found by Tietze, Guttmacher, and Rubin³⁵ in 1,727 planned pregnancies of fertile couples. Can these pregnancies be attributed to this procedure or to chance? Three of the patients, or 5 per cent, were thought to be pregnant at the time of the dilatation. All of us have seen the occasional patient who became pregnant after telephoning for an appointment, a situation also mentioned by Buxton and Southam.⁵ Cary⁷ had 4 pregnancies in 100 cases, or 4 per cent, that occurred incidentally to his other therapeutic efforts. Sharman³⁰ found that 10 per cent became pregnant after a simple bimanual pelvic examination. It would appear that a total pregnancy rate of 41.8 per cent after dilatation is also better than the 30 per cent found by Tietze and associates.³⁵ As indicated in Table IV, more than half of the conceptions occurred within one month after the dilatation. Table X shows that 42 per cent of Rubin's²⁹ patients also conceived within a month after the tubal insufflation. Rubin,²⁹ Cary,⁷ Stein and Leventhal,³³ Siegler,³¹ Gray,¹³ Buxton and Southam,⁴ and other observers have indicated that 25 per cent of the patients conceive after something is done. Tables III and IV revealed that 39 women in the 158 fertile matings, or 24.7 per cent, became pregnant within one month after the dilatation. Consequently, one must conclude that pregnancies occurring after cervical dilatation are not due to chance alone.

Summary

1. This is a study of 221 infertility patients of which 180 were treated by dilatation of the cervix in the office either as a primary procedure or as a follow-up measure to a hospital procedure. There were 158 fertile couples of which 66 or 41.8 per cent conceived after cervical dilatation.

2. Conception occurred within one month of the dilatation in 39, or 59 per cent, of the cases so that this procedure was given most of the credit. However, various other modi operandi are discussed.

3. There were 35 patients with cervical stenosis and 31 became pregnant after dilatation, showing the importance of this factor in infertility. However, 35 patients who did not have stenosis became pregnant after dilatation.

4. There was a group of 28 patients with refractory cases, of which 8 or 28 per cent conceived after simple office dilatation of the cervix.

5. Follow-up studies on the 66 couples that conceived originally after dilatation revealed 28 subsequent pregnancies, 7 of which followed a repeat cervical dilatation.

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Discussion

DR. S. LEON ISRAEL, Philadelphia, Pa.—During my reading of Dr. Javert's manuscript, I had constantly the feeling expressed by Freud's *déjà vu*,¹ an illusory sensation leading to the inference, "I have experienced all of this, just this way, before." It is the psychic mechanism that permits one the experience of entering a room for the first time but having the sensation of previously having been in that room. The *déjà* of this instance recalled that empiric cervical dilatation had been recommended before, even earlier than the published reports of Cary, Siegler, Birnberg, Sharman, and van Tongeren cited by Dr. Javert. Sims had emphasized the condition of the cervix as a cause of infertility.² He noted that the cervix was "conoid" in 175 of 218 (85 per cent) married women who had never borne a child; he attributed their barrenness to this. "For fertility," Sims wrote, "the cervix should be of proper size, form, and density." In fact if the cervix was too long, he amputated it; if it was too rigid, he cauterized it; and if it appeared stenotic he dilated it.

Although Javert refers to the fact that Buxton and Southam do not mention dilatation of the cervix as a therapeutic modality, he fails to note the reason for such a deliberate omission. They state, "The smallness or tightness of the external and internal ora is infrequently of importance in infertility problems."³ Inasmuch as the nature of the alleged cervical factor, as expressed by Buxton and Southam and as admitted by Javert, is a matter of opinion, doubt must exist concerning any therapeutic claims following its treatment. Surely, no cervix accepting an 11 Fr. dilator may be considered stenotic.

Selection of the major cause of infertility in any barren couple is difficult because there are no fixed standards to make the choice between several possible etiological elements an impersonal or automatic one. The decision is perforce arbitrary, conjectural, and personal some of the time, making it impossible to compare with accuracy the results of various therapeutic measures. An unprejudiced comparative appraisal is difficult to achieve in reports of sterility therapy because of diverse differences in age, nutrition, patterns of sexuality, and genotype of each partner. Moreover, as Dr. Javert has himself so earnestly contended elsewhere,⁴ the psychosomatic effects of being well attended medically or of being exposed to an arresting new maneuver cannot be evaluated accurately.

It is thus impossible to ignore the lightly glossed-over additional "therapy" included by Dr. Javert in his present series. Following the cervical dilatation, he recommends the knee-chest posture for intercourse. This unusual recommendation certainly requires consideration as a psychogenic stimulus, to say nothing of its possible enhancement of spermatozoal ascent. Ford and Beach⁵ pointedly state, "In some societies coitus occasionally occurs with the man standing behind the women while she bends over or rests on her hands and knees. The method is apparently confined to brief and sudden encounters in the woods." In our culture the knee-chest position for sexual intercourse is not usual and when actually prescribed by a physician, may exert an important influence. It may then be not, as Javert has indicated, that dilatation of the cervix is translated into a neural impulse, but rather that the acrobatic coital posture stimulates the hypothalamus.

Data derived from experimentally inflicted lesions, glandular implants, and clinical observations leave little doubt concerning the hypothalamic control of hypophyseal gonadotropic activity. It is important to bear in mind that the current acceptance of this physiologic mechanism implies two important concepts:

1. The anterior pituitary may no longer be regarded as "the conductor of the endocrine orchestra." In the language of Parkes,⁶ "The conductor is really just a marionette, activated by different strings pulled often simultaneously, by various members of the orchestra, as well as by members of the environmental audience. Clearly, this is no ordinary orchestra, and the fact that physiological cacophony is comparatively infrequent shows that the elaborate coordination is surprisingly effective."

2. The reflex arc that relates the genital sphere to the hypothalamic-pituitary center initiates neurohormonal stimuli which may either inhibit or, as the case may be, stimulate the pituitary's function. It would take us too far afield to recall here that this involves, at least sexually, hormones of the anterior and posterior pituitary lobes, for not only is gonadotropic function affected but there is also stimulation of myometrial activity because of oxytocin. The possibility of this hormone's enhancing uterine motility and, thus, spermatozoal ascent makes any reference to coital position, orgasm, and other emotional factors of great importance in evaluating the occurrence of pregnancy in a previously sterile woman.

In conclusion, may I summarize my reaction to Dr. Javert's paper by expressing disbelief in the efficacy of dilatation of what is admittedly a nonstenotic cervix to effect a wanted pregnancy. I would prefer to credit either the hypnotic effect of Dr. Javert's dynamic personality, as he wields the dilator, or the orgiastic ritual of the knee-chest posture.

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DR. ERLE HENRIKSEN, Los Angeles, Calif.—Adding anything of true value to Dr. Javert's paper would be idle boasting that, for me, the management of sterility holds no problems. Enough to say that I am in close agreement with his sensible approach.

If you read the papers, you are aware that the question of whether we should labor to increase the fertility curve or to depress the curve is receiving much space. One item describes, in unrestrained enthusiasm, the discovery of a new brew capable of making hens lay nothing but double-yolked eggs, mares foal nothing but derby winners, and women conceive with the first snap of a suspender button. The next column carries an epitaphic communication warning the world of the impending "fertility explosion." At the thirtieth Annual Meeting of the Aero-Medical Association in Los Angeles, attention was called to the effect of noise on fertility—hinting of the depressive effects of 115 plus decibels on oogenesis, spermatogenesis, and conception. It is distressingly clear that man is going all out to make the age-old physiologic act of bearing children most difficult. From review of many articles dealing with this problem, it is evident that there is much validity in that old Chinese proverb, "Never permit the complete truth to interfere with the beauty of thy word."

The admission by the author that his awareness of cervical manipulation is recent is indeed astonishing. To Dr. Javert and to all interested parties, I strongly commend the study of the articles, pertinent to this problem, written by the former presidents of this organization during those years starting with Marion Sims and ending with Isadore Rubin.

It has been said that "only the man familiar with the art and science of the past is competent to aid in its progress in the future." I am not certain the reader will necessarily become competent, but the logic displayed by these writers will instill him with a deep sense of humility—a wonderful attribute too often lacking in this modern day.

In 1868, Sims, our fourth president, wrote that "closure of the womb" was practically the only cause of sterility, advising dilatation of the cervix with sea-tangle or tupelo tents, and in the occasional case, division of the cervix with the hysterotome. His results were reported as excellent, and the cervix continues being dilated, straightened, amputated, and repaired.

I would like to see Birnberg's claim of a positive prolan response in 23 of 30 women, following cervical dilatation, deleted. His series was completely uncontrolled; his results never duplicated.

Little was said of the male factor, a factor of equal importance to that of the female. I have been impressed with the number of wives, pleading for help but refusing, in no uncertain language, to subject their mates to study. Some women seem to thoroughly enjoy bearing the onus of being the "sterile one." Certainly this attitude has extremely deep-seated implications. Then there is the widespread willingness to accept the inadequately established status of the sperm usually based on the number, the activity, and the examiner's idea of good health. It is readily admitted that we know more about the sperm than when it was discovered in 1677, or possibly when it was described as containing a miniature body in 1694. However, its relationship to fertility was not considered until 1853, and, despite the recent article by Carl Hartman, containing an illustration of the sperm not too unlike the blue print of a four-stage rocket, one need admit that there are many still so-called imponderables.

Then there are the questions regarding timing and frequency of the marital act. It is quite disturbing—the number of couples trying to base the timing of this important procedure on temperature graphs, the vast majority of which could not be interpreted by the most ardent thermometer enthusiast. Then we have the couple advised that they are having too frequent relations, who, in an honest effort to cooperate, shift to too few. What constitutes the ideal frequency? For guidance one can turn to the ancient legislative enactments of such as Zoroaster, Solon, or Mohammed all advising 3 to 4 connubial embraces each month, under normal conditions. Martin Luther edited a dictum that twice weekly was ideal. Balzac, displaying a keen insight of the fundamentals, held that it was necessary to play often if the odds against turning up the winning number were high. I go along in part with the sensible advice found in the Talmud, that "the frequency should be according to the social position and vocation." There are those who can run 2 miles each day without effort; there are those who tire after walking 2 blocks, but each in his own is normal.

The early investigators were either cagey or remarkably sensible, reporting their results in terms of poor, good, or excellent. Today, statistics are considered indispensable, be the number 2 or 2,000. After review of the literature, it is most difficult to reconcile the reported results with the innumerable forms of therapy. It is overwhelmingly evident that too many so-called imponderables, along with too many known partially known factors, never appear in the majority of the papers. Too often the investigator errs in interpreting coincidentals as synonymous with consequence. To return to the so-called "good old days" would, as the records now stand, be both sensible and worthwhile. This does not imply that I am not all in favor of continued investigation.

As for the cost, it is readily admitted that there are many worthwhile and highly ethical sterility clinics, but there is also an increasing number of self-styled sterility groups offering "packaged" examinations at prices often exorbitant. Some of the contracts differ little from the shady contracts offered by high-pressure salesmen in other fields, for, as the study progresses, the couple is surprised to find that they are being charged for so-called extras, not covered in the large print—a typical case of "the large print giveth, the small print taketh away." That the tests included in these package deals are not valuable is beside the point, but their promiscuous use without the slightest indication is

deserving of condemnation. The couples desirous of children constitute a most anxious and gullible group, and to sell a package, wrapped in gaudy terms and tied with hints of inevitable success, is certainly neither cricket nor honest. Surely the sage words of George Washington, the Father of our Country, to "labour to keep alive in your breast that little spark of celestial fire, conscience" is most applicable to this group of unfettered enthusiasts.

To explain many problems in the field of sterility, one need only turn to our extremely valuable but rarely appreciated ally, the husbandman. The cattleman well knows that there are good breeders and there are poor breeders. The good get pregnant easily and quickly, go through and deliver without difficulty, whereas the poor breeder has trouble both in getting pregnant and in carrying. We owe much to the field of animal husbandry and it would behoove us to study carefully its efforts and results. The lady of the farm handles the problem of poultry oogenesis according to the tenets of survival of the fittest. The poor egg layer ends up on the barbecue spit.

May I commend the author on his excellent work and at the same time request that we should not always take the credit for what nature frequently accomplishes, despite interference by man which is often both ill-advised and bungling. It is well to be humble with the awareness that women have been conceiving from time immemorial and that each era of the past, self-satisfied with its acclaimed knowledge, has been contented with its *modus operandi* and its results.

THE GLANDULAR STRUCTURES OF THE CERVIX UTERI DURING PREGNANCY*†

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THE prevalent concept of the anatomy of the cervix uteri during pregnancy is based on the classic description by Stieve,¹ who attributed the fundamental changes to a hypertrophy and dilatation of tubular racemose glands. Since it has been shown that the cervical mucosa is not composed of this type of gland but of a complicated system of compound clefts,²⁻⁴ it has seemed desirable to review this subject by use of other methods of investigation besides the study of conventional histologic preparations. The present report is concerned with the nature of the glandular structures of the cervix during various stages of gestation as determined by photographic techniques, plastic reconstructions, and serial microscopic sections.

Material and Procedures

The 39 specimens of pregnant uteri available for this study were obtained by total hysterectomy, and although there were several indications for this operation none were for primary disease of the cervix. Eleven were from the first trimester of gestation, 5 from the second, 4 from the third, and 19 were from term pregnancies. Five additional specimens were obtained from 2 to 4½ months after delivery and 2 were from hysterectomies performed immediately postpartum for hemorrhagic complications. All patients were multigravidas except 2, one in the second trimester and one at term, who were in their first pregnancy; they varied in age from 20 to 44 years. From one to 6 longitudinal sections of the cervix were prepared from each specimen and after fixation in formalin and mounting in paraffin they were cut at 10 μ and stained with hematoxylin and eosin.

The surface markings of the cervical canal were studied in 11 specimens at term either with the naked eye or under a stereoscopic microscope or magnifying glass. Drawings or appropriate photographs at a magnification of 3 or 4 were made. Although they were originally fixed in formalin, it was found desirable to immerse these specimens in 95 per cent alcohol for several days before study.

A segment of 7 specimens was prepared in the manner previously described to obtain thick cleared sections for observation under a stereoscopic microscope.² This consisted of making freehand sections approximately 0.5 to 1.0 mm. in thickness, clearing in xylol and oil of wintergreen, and staining lightly with Mayer's hematoxylin, 1 per cent.

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The panoramic views of the cervix given in Fig. 6 were prepared in various ways but the method finally adopted consisted of a number of distinct steps. First, 3 or 4 photomicrographs were taken of adjacent fields of the cervical canal to cover a distance of one inch at a magnification of $\times 15$. The photographs were then cut and pasted side by side so that outlines of the epithelial structures could be traced onto sheets of cellulose acetate. They were then transferred to paper and a black background painted in with India ink. This was now rephotographed and a print made of a light gray color. The outlines again were drawn in with black ink and the final photographs made. The original preparations were 15 inches wide, but they represented an actual distance of one inch in the microscopic sections, and they were reduced to the desired size in the prints used as illustrations.

The serial sections were cut $8\ \mu$ thick and stained with hematoxylin and eosin. Six series totaling 2,016 sections were prepared; 5 were cut longitudinally to the cervical canal, and the sixth transversely. The first series was composed of 274 sections, and was from a specimen obtained at approximately the fifth week of gestation. The exact date is not known because the patient had had irregular bleeding, but the fetus measured 1.5 cm. in length. The second series, 276 sections, was from the tenth week; the third, 376 sections was from the thirteenth week; the fourth and fifth, 225 sections in each case, were from a term pregnancy. The sixth series, 640 sections, was cut transversely from a specimen of term pregnancy.

In addition to their availability for study, the serial sections were used for preparing plastic reconstructions of the cervical epithelium. The method used was originally described to me by Dr. Arthur T. Hertig, but it has been in use for many years by the Department of Embryology, Carnegie Institution of Washington.⁵ The details of the modified procedure employed in these studies were given in a previous report.³ The basis of the technique consisted of drawing outlines of the epithelial structures from photomicrographs on sheets of clear plastic material (cellulose acetate). These transparent plastic enlargements were then arranged in serial order to give a three-dimensional effect. A distinct advantage of using these outlines is the elimination of unnecessary detail although basic accuracy is not sacrificed, and, with special care in superimposing the anatomic structures, the continuity of the clefts is clearly demonstrated. Although the plastic transparencies may be separated by a scale distance to preserve the accuracy of the third dimension, it has not been found necessary to employ separators in this study. During pregnancy the cervical clefts with their secondary clefts, tunnels, and projections are relatively large structures so that it has been a simple matter to follow them by using every third to sixth section. The illustrations of the outlines used in this report are as much as 18 to 36 sections apart. A number of such plastic reconstructions were prepared from the 6 series of serial sections, and they represent early and late stages of pregnancy.

The Cervix at Term

During the course of gestation the cervix undergoes striking changes which affect all its individual components and result in the transformation with which all clinicians are familiar. As term approaches, it becomes shorter and broader and very soft; the external os, at least in multigravidas, readily admits the examining finger. The tremendous increase in vascularity due to the formation of numerous new blood and lymph vessels and dilatation of arteries and veins, is greatly responsible for these alterations, but there is likewise a profound modification of the character of the lining mucosa.

The columnar cells lining the cervical canal and its clefts and tunnels show signs of tremendous activity both in growth and in secretory function.

The individual cells increase in size, becoming taller and broader, and as they increase in number they are heaped up and stratified. A direct result is the formation of irregular projections on the surface and along the walls of the clefts, and, because of their number and the pressure of surrounding tissues, they come to form secondary clefts and fingerlike processes which in turn result in spaces filled and distended by mucous secretion. These changes become apparent very early in gestation but an increase in the vertical depth of the mucosa is not prominent until the third trimester. Actual measurements are shown in Fig. 1, and they compare closely with those given by Stieve.¹ In this diagram each dot represents the mean of 5 measurements taken on one

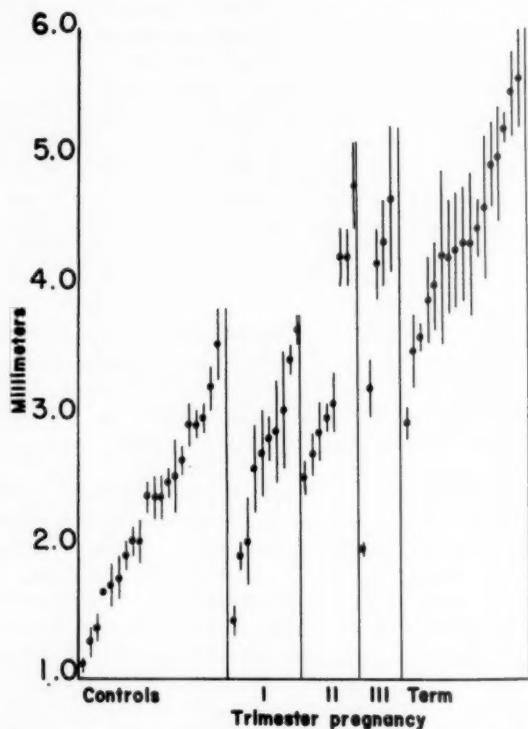


Fig. 1.—Measurements of the depth of the cervical mucosa obtained from microscopic preparations with an eyepiece micrometer. Each dot represents the mean of 5 measurements taken from one slide and the vertical lines through the dots show the spread. The marked increase in the depth of the mucosa during pregnancy is apparent during the late second trimester, in the third trimester, and in the newborn baby at term.

microscopic slide and the vertical lines through the dots show the spread. The dots are arranged from low to high in each category, and it is readily seen that there is a great variation in each case so that an average figure would not have very much significance unless many more cases were available. Nevertheless the distribution of these figures is instructive. In the normal nonpregnant cervix the depth of the mucosa varied from 1.2 mm. to 3.5 mm. During the first trimester of pregnancy the spread was similar but a greater number were noted between 2.5 mm. and 3.5 mm. In the second trimester the figures fell into two groups, some between 2.5 and 3.0 mm. and others between 4.0 and 5.0 mm. Only 5 sets of measurements were made in the third trimester and 3 were between 4.0 and 5.0 mm. The maximal depth of the mucosa was attained in the cervix at term and varied from 3.0 to 6.0 mm. Since these measurements were taken from microscopic preparations the actual size in fresh specimens obviously would have been considerably greater.

The active secretion of the cervical cells results in changes which are apparent on gross examination. The canal is filled with a clear, jellylike, mucoid substance which corresponds to the "mucous plug" seen in many species. The surface markings are very prominent and the larger rugae and depressions not only can be seen readily with the naked eye but they are easily photographed. The classic longitudinal folds with smaller accessory folds running obliquely or transversely to form a regular arrangement of "plicae palmatae" or "arbor vitae" are nonexistent as in the nonpregnant.⁴ There is no attempt at arborization but instead there are numerous folds and grooves of varying size which course in all directions (Fig. 2). Some extend throughout the whole

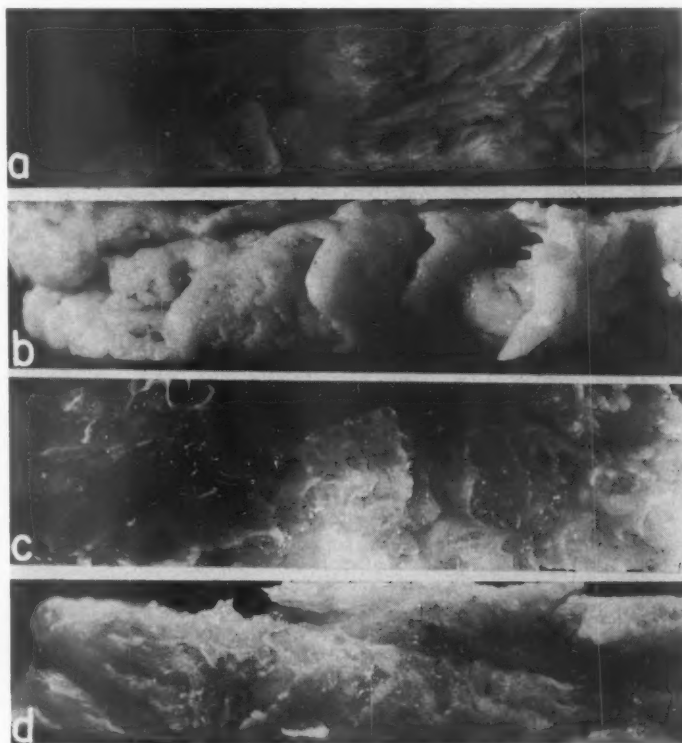


Fig. 2.—Photographs of strips of the cervical canal after fixation in formalin and clearing in alcohol. There is no pattern of arborization but many irregular prominences and grooves course in all directions. In pregnancy at term (*c*, *d*), characteristic findings are circular or oval compartments which appear in the photographs as small pits. Section *a* is from an early pregnancy of about 5 weeks, and *b*, *c*, and *d* from term gestations. (Magnification, *a*, *b*, and *c*, $\times 4$; *d*, $\times 3$; reduced $\frac{1}{3}$.)

depth of the mucosa while others are shallow, and they run in straight or curved lines without following any regular pattern. They form round or oval cuplike depressions, some minute and others relatively large, which in the fresh specimen are filled with tenacious mucous secretion. These depressions are found to a lesser extent in the nonpregnant uterus but they are particularly characteristic of pregnancy and, as shown in Fig. 2, *C* and *D*, they occur in great numbers in the cervix at term. This gross appearance fits in with the histologic picture described later, but it must be emphasized that they vary to such a degree that no 2 specimens follow the same pattern. It is of interest that the formation of these crypts or pits is reminiscent of the description of "mucous follicles" given in 1855 by Tyler Smith⁶ as a normal constituent of the cervical mucosa.

The study of vertical thick (0.5 to 1.0 mm.), cleared sections, whether cut longitudinally or transversely to the canal, presents an irregular arrangement of the cervical mucosa into oval or round compartments (Figs. 3 and 4). They vary in size, some extending the whole depth of the mucosa while others are smaller and appear stratified one above the other and separated by smooth partitions. They correspond to the pits or crypts seen in the surface markings and to the histologic clefts and tunnels found in serial sections. They are a constant finding in the cervix at term and were demonstrable in some 50 cleared sections prepared from 7 term specimens. This observation has been made many times from conventional microscopic sections and it has been de-

Fig. 3.

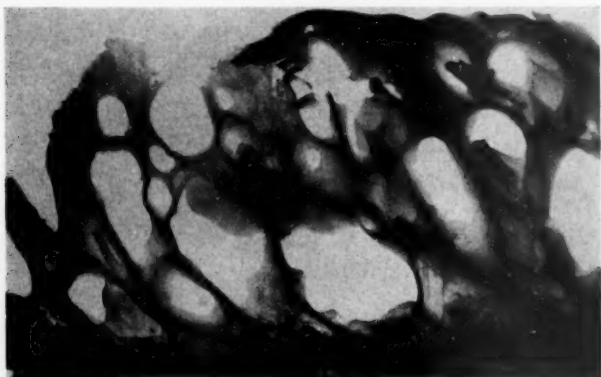


Fig. 4.

Fig. 3.—Section of the cervical mucosa, from a pregnancy at term, which was cut about 0.5 mm. in thickness and cleared in zylol and oil of wintergreen. ($\times 6$, approximately; reduced $\frac{1}{3}$.)

Fig. 4.—Section of the cervical mucosa, from a pregnancy at term, which was cut about 0.5 mm. in thickness and cleared in zylol and oil of wintergreen. ($\times 5$, approximately; reduced $\frac{1}{3}$.)

scribed as a "honeycomb" appearance. Actually, however, the structures vary greatly in size and do not assume a hexagonal shape so that a honeycomb is not a very apt metaphor.

Cervical Clefts During Pregnancy

The transformation of the epithelium of the cervical canal into the complicated arrangement found in pregnancy at term follows an evolutionary trend which begins early in fetal life.² At first the mucosa is thrown up into multiple folds which course in longitudinal, transverse, and oblique directions and, with

the growth of the surrounding tissues, they become compressed to form larger and smaller grooves. Under the influence of maternal hormones, the cervix of the newborn baby undergoes radical changes which involute after birth, and, with the menarche and the onset of menstrual life, renewed activity sets in. As pregnancy supervenes later in life the cervix uteri once more reacts to an intense stimulus which brings about changes which are similar but far more complicated than those found in the newborn.

Since it has been accepted that the cervical mucosa is composed of tubular glands which descend from the surface and branch in a racemose arrangement, it was inevitable that Stieve and his successors should explain the pregnancy changes as resulting from a hypertrophy and dilatation of such glands. There

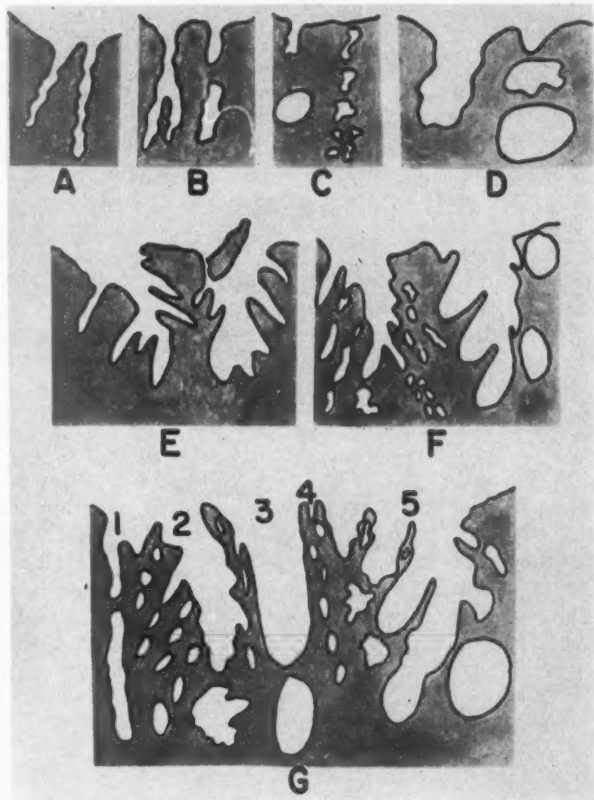


Fig. 5.—Diagram illustrating the basic clefts of the cervical epithelium and the modifications which they undergo during the course of gestation.

are, however, no tubular glands of this type in the cervix at any age period and this conclusion has been reached by means of a number of different techniques as well as by the study of some 5,000 serial sections and a considerable number of plastic reconstructions.^{2, 3, 4} Obviously, these observations disprove the conclusions reached in my contribution before this Society in 1947 that "adenomatous proliferation of the glands" is an important factor in the production of cervical erosions.⁷

The basic epithelial structure is not a tubular gland but a cleft or groove as shown in cross-section in Fig. 5, A. As the name implies, it is simply a cleft which runs in an oblique, transverse, or longitudinal direction, and, contrary to past beliefs, it definitely is not a tubular gland which descends from the

surface to divide in a racemose pattern. At times it is relatively shallow but often dips down through the whole thickness of the mucosa. The larger correspond to the deep grooves seen in the surface markings but there are innumerable other shorter and shallower grooves in between the large structures. The actual length of the shorter clefts usually varies from 100 μ to 1.0 or 2.0 mm. but they may be as long as 1.0 or 2.0 cm., judging from the dimensions of the surface markings.

It is in the modifications which the basic clefts undergo that one must look to explain the appearance of the cervical epithelium in a term pregnancy. However complicated this process may seem, it is based on a few fundamental changes which are shown in the diagrams of Fig. 5. They consist of (1) tunnelloid extensions, (2) exophytic processes and secondary clefts which also may form tunnels, and (3) hypertrophy and dilatation of all the structures due to hyperplasia and hypersecretion of the mucosal cells.

1. *Tunnels*.—An important characteristic of the clefts is their tendency to become occluded so that an area becomes pinched off and continues as a tunnel or blind tube (Fig. 5, B and C). These occlusions may be multiple so that a considerable number of smaller tunnels eventuate. The only exit to the surface that the tunnels have is their connection with the open cleft, and often they extend for longer distances than the open cleft from which they originate. Of importance is that these tunnels *always* course in the same general direction as the parent cleft and this finding in itself is a strong argument against any theory that they may be a type of invagination. At times a segment of cleft becomes occluded for a short distance and then reopens again, leaving a tunnel connecting what now appears to be two short clefts. Tunnels occur in relatively small numbers in the newborn cervix and they are usually absent in premature babies and scant in children so that they are a characteristic chiefly of the adult organ.

2. *Secondary Clefts*.—The second important feature of the compound cleft is that with an overgrowth of the lining cells there result excrescences which vary in size and shape, and these again lead to the formation of secondary clefts which extend laterally or downward (Fig. 5, E). These secondary clefts themselves develop tunnels and they may also form further clefts and projections (Fig. 5, F). The resultant structure obviously may become exceedingly complicated and this arrangement can be correctly interpreted only by a painstaking analysis of outlines traced on sheets of plastic material. It is not unusual to see a microscopic field with an endless number of oval and round spaces representing cross sections of tunnels but which formerly were accepted as segments of tubular glands coursing down from the surface. On serial section and with reconstruction, however, it is easily demonstrated that they are not part of vertical tubular glands but run *more or less parallel to the surface* and eventually join with a cleft.

3. *Growth and secretory activity*.—The enhanced cellular activity during pregnancy not only leads to the production of many exophytic growths from the walls of the clefts and secondary clefts, but with hypersecretion there is a widening of the clefts and a distension of the walls of the tunnels (Fig. 5, D, F, G). This is an active process so that the lining cells do not necessarily become low and cuboidal unless a tunnel becomes blocked and it is then the equivalent of a nabothian follicle. This process is responsible, to a considerable degree, for the increase in depth of the mucosa and it results in a thinning out of the stromal walls between the clefts and tunnels. The resultant protrusion of fingerlike processes which enclose spaces filled with secretion is typical of the cervix at term and is demonstrated in Figs. 5, F and G, 6, C and D, 8, 9, and 10.

The changes of the cervical mucosa at term are summarized in the diagram given in Fig. 5, G. Five clefts are shown from left to right. The first is a

simple cleft with an occlusion at a point about one-third down from the surface and leaving a tunnelized portion beneath it. The next is a large cleft and tunnel with secondary clefts with their tunnels. The third is a single large cleft and tunnel, and it is separated from the second cleft by a fingerlike protrusion. The fourth cleft is represented by a small dip in the surface and 6 small tunnels directly below. The fifth cleft, on the right, shows a distended cleft with secondary clefts, tunnels, and fingerlike processes. The large, oval space in the lower

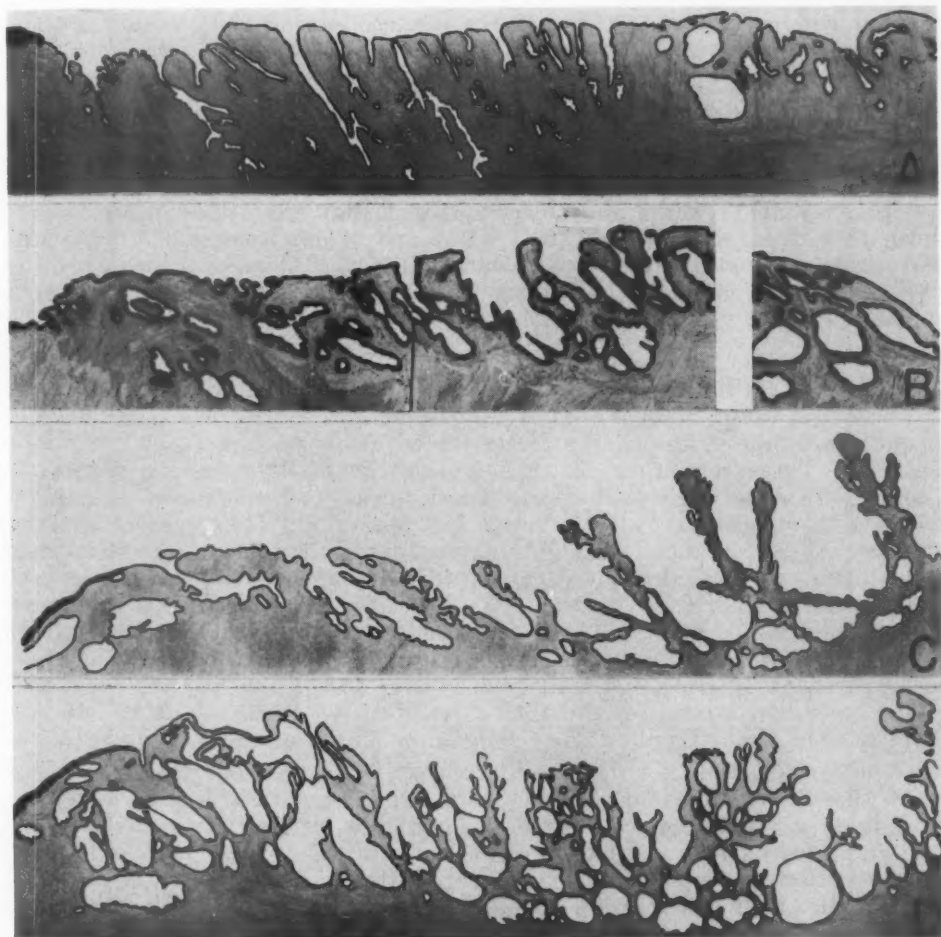


Fig. 6.—Panoramic views of the cervix uteri obtained by making tracings from photographs of microscopic sections taken at a magnification of $\times 15$. The width of each diagram covers a distance of one inch in the original slide. *A* is from a nonpregnant patient; *B* from a pregnancy at 13 weeks; *C* at 35 weeks; and *D* from a pregnancy at term. (Reduced $\frac{1}{4}$.)

right-hand corner could be the equivalent of a nabothian follicle provided the cells were of a low cuboidal type due to mechanical distension of an occluded tunnel. Although Fig. 5 is a diagram prepared to illustrate these various changes, it should be understood that it is based on actual observations made with plastic reconstructions.

Evolution of Clefts During Pregnancy

Although this series is small it is representative of all the months of pregnancy, but it has been impossible to establish definite criteria which could be

used to recognize any particular stage beyond the wide variations of early and late gestation. This is due not only to the great variations and degrees of changes which are seen in the cervix at comparable periods but also to the marked differences found in sections from the same specimen. This observation is in keeping with the variability noted in the measurements of the mucosa. It is, however, possible to recognize changes characteristic of pregnancy as early as 6 to 8 weeks by the increased secretory activity of the cells as well as by the appearance of larger clefts with numerous distended tunnels and projections into the spaces of the clefts. During the second trimester these changes are more pronounced and some of the features seen at term become apparent, but as a rule the exaggerated picture of the pregnant cervix is not fully developed until the last 3 months. Fig. 6 presents panoramic outlines from 4 specimens, and each covers an area one inch long in the lower part of the cervical canal and just above the external os with its squamous epithelium. The first, *A*, is from a nonpregnant patient. The second, *B*, is from a pregnancy at 13 weeks and shows enlarged cleft spaces and tunnels. The third, *C*, is from a pregnancy at 35 weeks and gives a remarkable demonstration of the production of "compartments" filled with secretion which can be so easily recognized on gross inspection. Fig. 6, *D* is from a pregnancy at term and shows all the changes which were discussed in the previous section.

Plastic Reconstructions

The most useful method of studying the cervical mucosa has been the usage of outlines of the epithelial structures traced on sheets of transparent plastic material. These reproductions are simple to manipulate, can be marked as desired with colored grease pencils, and, accurately superimposed, they allow the various structures to be followed and analyzed. They present a three-dimensional view of the clefts and associated formations, but it has been found impossible to prepare any type of flat drawing from them which could be used to illustrate a report such as this one. For this reason it is necessary to reproduce sequential drawings from the serial outlines. Although they give a general perspective of the clefts and tunnels, they are necessarily far apart because of lack of space, and consequently they do not give all the finer details of the secondary structures and their development. Figs. 7, 8, 9, and 10 give three examples, one from an early pregnancy and two from cervixes at term.

The first example is a longitudinal section of the cervix from an area just above the squamocolumnar junction (Fig. 7). The patient was 25 years old, gravida v, and in a very early stage of pregnancy, probably 5 weeks since the embryo measured 1.5 cm. A total of 274 serial sections were made and plastic reconstructions were prepared by using every fourth section. In this diagram every sixth outline is reproduced so that it represents every twenty-fourth section, and this sequence is indicated by the numbers in the corners of each drawing. The width of each diagram corresponds to 5.1 mm. and the outlines from 0 to 168 cover a distance of 1.3 mm. The maximal depth of the mucosa noted in this series was 3.1 mm. An idea of the measurements also may be obtained by noting that the open part of cleft *V* in diagram 72 is 2.8 mm. in depth. At this stage of pregnancy there is a considerable proliferative activity of the columnar cells so that the clefts have many irregular projections and numerous tunnels are formed. In this diagram, 4 areas are marked off by dotted lines and are indicated by the letters, *O*, *V*, *X* and *Z*. Each contains interconnecting structures which can be superimposed and followed from one section to the next. The area denoted by *Z* contains a number of smaller clefts and a large one which is seen in outline 144. The other 3 *O*, *V* and *X*, however, each contain one major cleft and tunnels with secondary clefts and tunnels. They

are present in the first section (outline 0) and continue for various distances until they finally disappear with only vestiges of small tunnels. Cleft X ends with section 144, and consequently it runs for a distance of 1.15 mm. as shown in the diagram. Clefts O and V continue beyond the limits of the diagram, O terminating with section 212 for a distance of 1.69 mm. and V with section 192 or 1.53 mm. These measurements are easily obtained since each section is $8\ \mu$ in thickness and it is merely a question of multiplying the number of the sections by this figure.

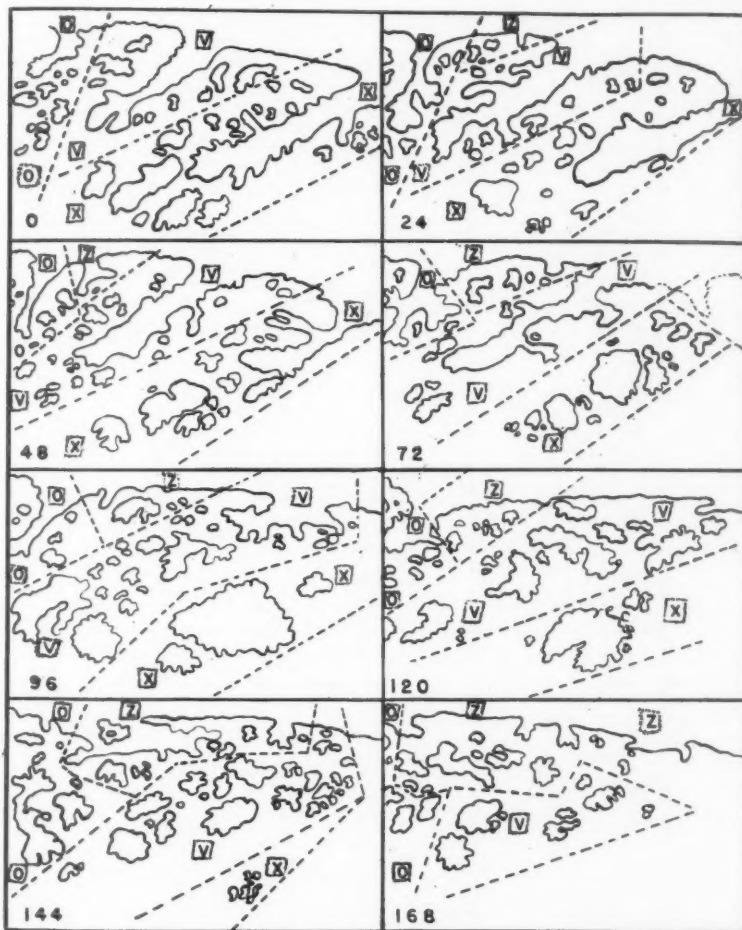


Fig. 7.—Tracings made on plastic material of the epithelial structures of a cervix uteri from a pregnancy at 5 weeks. As indicated by the figures in the lower left-hand corner, they represent every twenty-fourth microscopic section. The dotted lines separate areas designated as O, V, X, and Z, which are described in the text. ($\times 12$; reduced $\frac{1}{2}$.)

The 18 outlines given in Figs. 8 and 9 are from a specimen of the cervix uteri at term from a patient, gravida iv, 29 years of age. They were selected from 640 serial sections prepared from a transverse segment of the middle third of the cervical canal. The first 3 outlines are 36 sections apart and from then on they are 18 sections apart as indicated by the numbers in the upper right hand corner of each diagram. The 2 main diagrams (Figs. 8 and 9) cover a distance represented by 342 sections or 2.7 mm. The dimensions of the first

outline 0 are 6.8 mm. in height and 5.1 mm. in width. The main interest of this series is that it represents the evolution of well-defined secondary clefts and tunnels which are numbered from 1 to 12 and which are part of a large or main cleft, indicated by the letter A. In sections 0 to 162 only one side of this cleft is visible but in 180 and onward it becomes smaller and both sides are seen. This main cleft as well as its secondary structures was traced beyond the limits of these diagrams through 510 sections, a distance of 4.08 mm. where it finally ends with tunnels that originated from secondary cleft 10. Cleft 10 begins as a

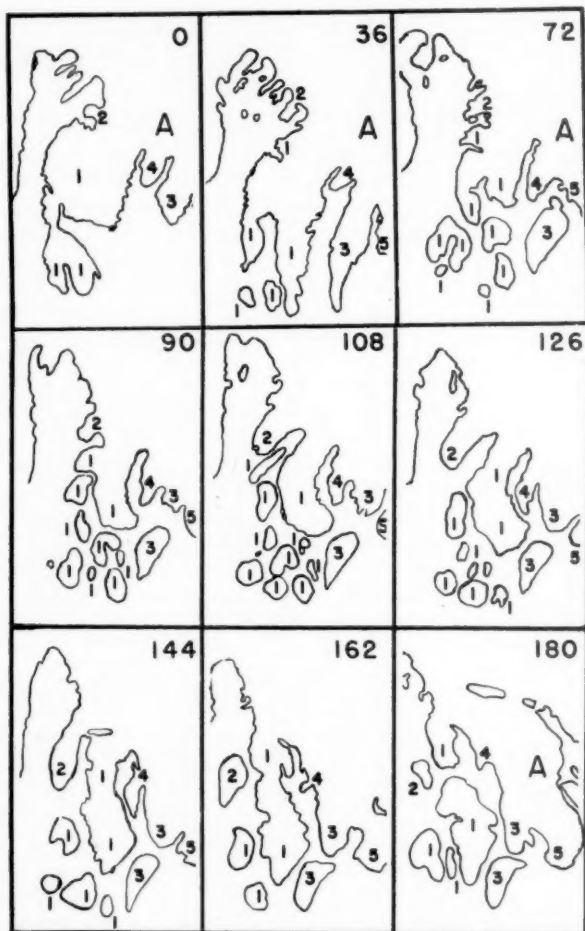


Fig. 8.—Outlines used in a plastic reconstruction of an area of the cervix uteri from a pregnancy at term. The figures in the upper right-hand corners give the sequence of the microscopic sections from the first tracing, labeled 0. The distance covered by Figs. 8 and 9 is consequently of 342 microscopic sections, or 2.7 mm. The letter A indicates a major cleft while the numbers identify secondary clefts arising from it. ($\times 7$; reduced $\frac{1}{10}$.)

small tunnel in outline 234 and then appears in outline 252 as an open cleft which increases gradually in size eventually to end in small tunnels after a course of 2.3 mm. The diagrams 0 to 342 show the development of secondary cleft 1, which begins as a bifurcated open structure in 0 and, after 216, is reduced to small tunnels which terminate in outline 342, thus being apparent for a distance of 2.7 mm. Cleft 6 begins in the section 198 and ends at 504, beyond these diagrams and for a total distance of 2.5 mm. The fingerlike processes

which take part in the origin of various clefts and the open areas which contain mucoid secretion and are so characteristic of the cervix of pregnancy are seen in all these outlines.

The third diagram, Fig. 10, is at a somewhat larger magnification and presents 5 outlines from longitudinal sections of a cervix at term. The patient was gravida iii, age 36 years. In this case a dark background was painted in in order to show more details. The width is 0.8 mm. and the total distance covered by the 5 outlines is 0.768 mm. In the original section from which outline 0 was

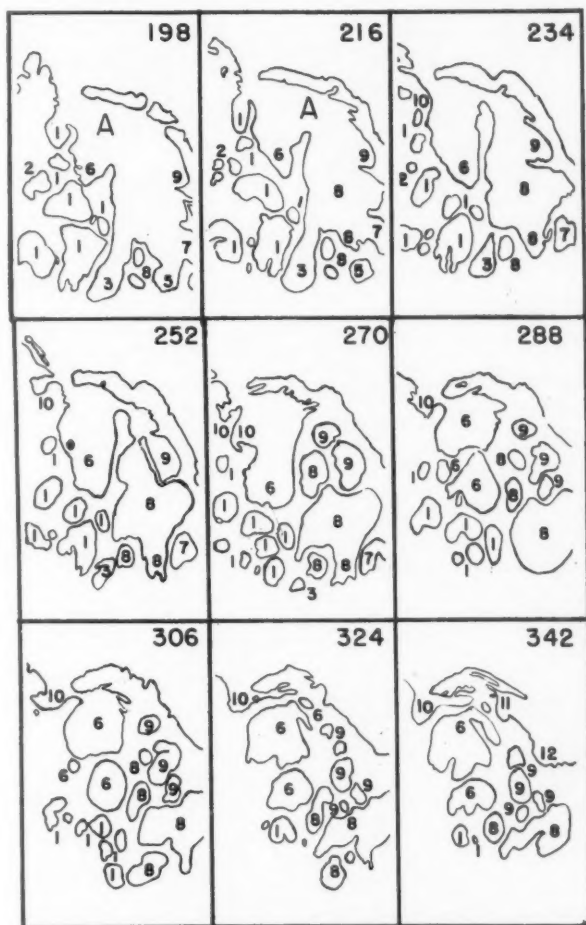


Fig. 9.—Continuation of Fig. 8. ($\times 7$; reduced $\frac{1}{4}$.)

obtained the clefts were not very prominent but two are indicated by the letters O and V while cleft X gradually evolves from tunnels seen in outlines 0 and 24. Numerous smaller clefts are visible in the walls of these larger clefts and from them arise the many tunnels which appear throughout each diagram. These structures are all secondary to a still larger cleft, the lumen of which lies to the left, and as a consequence they run in an oblique direction.

The Postpartal Cervix

Stieve¹ maintained that during parturition pressure from the products of conception forces the whole cervical mucosa out of the canal leaving only a few

glandular rests. Following this massive destruction, however, he found that there is rapid healing so that by the ninth day post partum the mucosa is well developed with folds and some glands.

The material available for this study is inadequate to reach any conclusions on the postpartal cervix. The 2 specimens obtained immediately after labor show evidence of trauma and extensive hemorrhages into the substance of the cervix, but it was impossible to gain any information regarding the mucosa. The 5 specimens from patients 2 to 4½ months post partum are

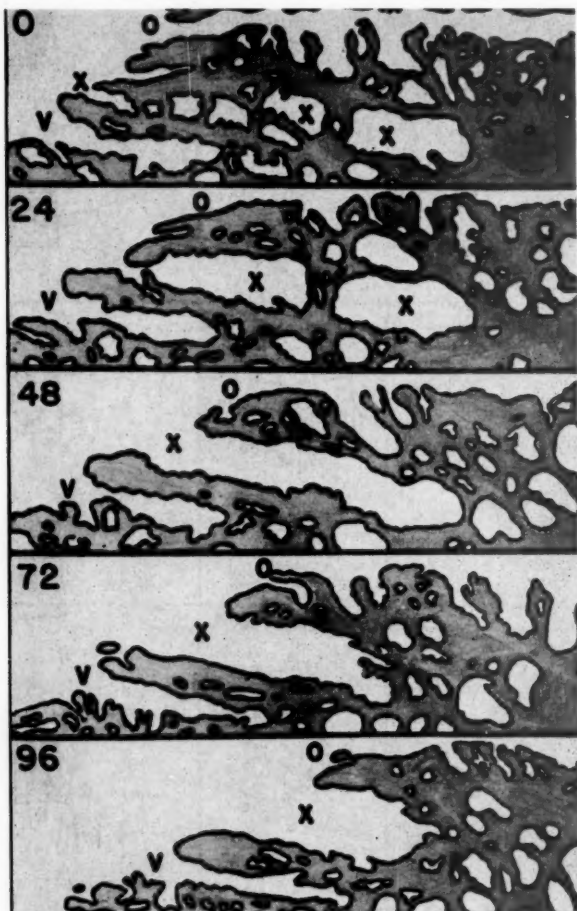


Fig. 10.—Five outlines, 24 sections apart, from an area of the cervix uteri in a term pregnancy. Three clefts are identified by the letters O, X, and V. ($\times 14$; reduced $\frac{1}{4}$.)

identical to the normal adult cervical mucosa with glands and tunnels. It was not possible to recognize any changes which could be associated with a previous pregnancy and labor.

The trauma associated with dilatation of the cervix and the passage of the baby seems bound to damage the delicate structures which develop in the epithelium of the cervical canal during the course of gestation. However, Revoltella⁸ and Glass and Rosenthal⁹ could not corroborate the claim that there is an extensive denudation of the epithelium in the postpartal cervix and further observations should be made from specimens obtained at different stages of dilatation of the cervix. Smith⁶ believed that the rugae of the cervix

allow for the distension which occurs during dilatation and prevent extensive mucosal lacerations. He stated that "during pregnancy the rugae are enlarged and loosened; but when the os uteri is fully dilated in labor, the mucous membrane of the cervix may be felt perfectly smooth, no doubt from the unfolding of the rugous mucous membrane. In this respect . . . the rugous arrangement of the mucous membrane of the cervix uteri may be compared to the rugous arrangement of the mucous membrane of the vagina." The fate of the cervical mucosa during labor will not be solved easily, but in this regard attention should be directed to the spontaneous involution which occurs in the cervix of the newborn.² In this case there is no trauma involved and the mucosa returns to a normal status with the cessation of the stimulus derived from the maternal and placental hormones.

Summary

This investigation was based on 39 specimens of the cervix uteri at different stages of gestation. They were studied by various methods including special photographic techniques, thick cleared sections, conventional microscopic preparations, serial microscopic sections, and reconstructions on sheets of transparent plastic material.

As in the nonpregnant state the cervical mucosa during pregnancy does not contain tubular racemose glands. The basic structure is a groove or cleft of varying dimensions which undergoes certain fundamental changes: (1) by a process of occlusion it forms tunnels which course more or less parallel to the surface and in the same general direction as the parent cleft; (2) the walls of the cleft may develop exophytic growths which project into the lumen and contribute to the formation of secondary clefts; (3) the secondary clefts themselves may form extrusions and tunnels; (4) as a result of enhanced growth and secretion during pregnancy these fundamental structures become greatly increased in size and number. The thickness of the cervical mucosa in term pregnancies measured from 3.0 to 6.0 mm. as compared to 1.2 to 3.5 mm. in the nonpregnant state.

My thanks are due Miss Ann Krepelka for her technical assistance and Mr. W. E. Renner for the photomicrographs.

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Discussion

DR. JOHN B. MONTGOMERY, Philadelphia, Pa.—The anatomic details that Dr. Fluhmann has presented to us are based upon his previously proved concept that the classically described compound racemose glands opening on the surface by a small duct

do not exist in the endocervix. He has shown clearly that the endocervix consists of a maze of clefts and grooves with secondary clefts and tunnels, the latter occurring most prominently under intensive hormonal stimulation.

His studies indicate that the complicated structures that develop during pregnancy come about by 3 fundamental changes in the basic folds or clefts in the endocervix which were developed primarily during intrauterine fetal life. First, tunnellike extensions develop from the clefts or grooves by a process of occlusion, and they often extend beyond the limits of the parent cleft. Although Dr. Fluhmann does not explain the mechanism of this, one might assume that it is akin to the process of budding by which gland structures generally are known to push out into surrounding tissues. Second, exophytic processes from the surface of the clefts and grooves form secondary clefts and apparently extend to the point of partial or complete occlusion of the cleft or groove. Tunnels are formed by this mechanism also. Thus, the picture is further complicated by hypertrophy and dilatation of all the structures due to hyperplasia and hypersecretion.

Dr. Fluhmann has shown that the only exit of the tunnels is their connection with the parent cleft and that they always course in the same direction as the parent cleft. It would seem therefore that each primary cleft with its secondary clefts, grooves, and tunnels might be regarded as a functional unit, somewhat akin to the classically described compound racemose glands, but more efficiently constructed from the standpoint of the effective secretion and flow of cervical mucus with its rapidly changing chemical and physical characteristics. The mechanism of the free flow of mucus from the previously described racemose glands of the cervix has never been clearly understood. Dr. Fluhmann's convincing demonstration of the true microscopic anatomy of the endocervix begins to clarify this point. It provides us with a structural detail that should lead to a better understanding of the function and the pathologic anatomy of the endocervix.

DR. FLUHMANN (Closing).—This study has dealt only with the anatomy of the cervix, and the extent to which the demonstration of clefts and tunnels will modify our concepts of function is only a matter of conjecture. The next step inevitably leads to the problem of the dissemination of carcinoma along these structures. The manner in which areas of so-called focal hyperplasia or adenomas are formed also calls for investigation and from preliminary studies it appears that they are artefacts resulting from secondary clefts and tunnels.

THE EFFECTS OF PREGNANCY AND LABOR ON THE RESPIRATORY PATTERN OF THE NEWBORN INFANT—SUBSEQUENT MORBIDITY AND MORTALITY*

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(From the University of Kansas Medical Center)

THE simple yet basic observation¹ that one can give a highly accurate prognosis with respect to survival for most liveborn infants, excluding only those with gross developmental anomalies, within a few hours after their birth has been very impressive to us. From the smallest babies (1,000 grams) on up, we have observed no deaths when the breathing pattern (Fig. 1) was in Group I or Group II.†

All the deaths in the first weeks have been in Group III, which must therefore be considered as an abnormal breathing pattern. Thus, it is evident, one can, by simply counting and charting the trend of the resting respiratory rate beginning at birth, (1) give a reasonably accurate prognosis as to survival; (2) relieve nursing personnel of the necessity of close and detailed attention to those infants with normal breathing patterns, and (3) concentrate their attention on the smaller percentage (Table I) that have an abnormal pattern.

TABLE I. INCIDENCE OF GROUP III BREATHING

	TOTAL PATIENTS	GROUPS I AND II BREATHING	GROUP III BREATHING	
			NO.	%
<i>Term Infants.—</i>				
Vaginal delivery	216	205	11	5
Cesarean section	155	138	17	11
<i>Prematures.—</i>				
2,000-2,495 grams	416	356	60	14
1,500-1,995 grams	144	80	64	44
1,000-1,495 grams	51	12	39	76
<i>All Prematures.—</i>				
Vaginal delivery	611	448	163	27
Cesarean section	34	16	18	53

As might be expected, full-term infants, delivered vaginally, seldom exhibit this type of abnormal breathing. This abnormal pattern is found twice

*Presented at the Eighty-second Annual Meeting of the American Gynecological Society, Hot Springs, Va., May 21-23, 1959.

†Group I starts with a "normal" respiratory rate of about 40 per minute and maintains approximately that level thereafter. Group II starts with a considerably higher rate but decreases promptly—or somewhat more slowly—to approximately 40 per minute eventually. Group III, on the other hand, may start slowly, about normal, or somewhat higher, but shows an increase in the first few hours (roughly 15 per minute).

as often among babies delivered by cesarean section, whether they are full-term or premature. Prematurity results in progressive proportions of abnormal breathing as the degree of prematurity increases. (It should be repeated that the infant, no matter how premature, that breathes normally has an excellent prognosis.)

The significance of this abnormal pattern of breathing, prognosiswise, is shown in Table II. The fact that there were no deaths in term infants, whether they were delivered vaginally or by cesarean section, is almost certainly fortuitous.* We know from other material available to us that death does occur at term in some babies with Group III breathing. It would seem that the chance of death following cesarean section would be at least double the chance of death following vaginal delivery, as the incidence of Group III breathing is twice as great (Table I). Obviously this presentation must be regarded as a preliminary report since the number of cases studied is too small to justify anything like final conclusions.

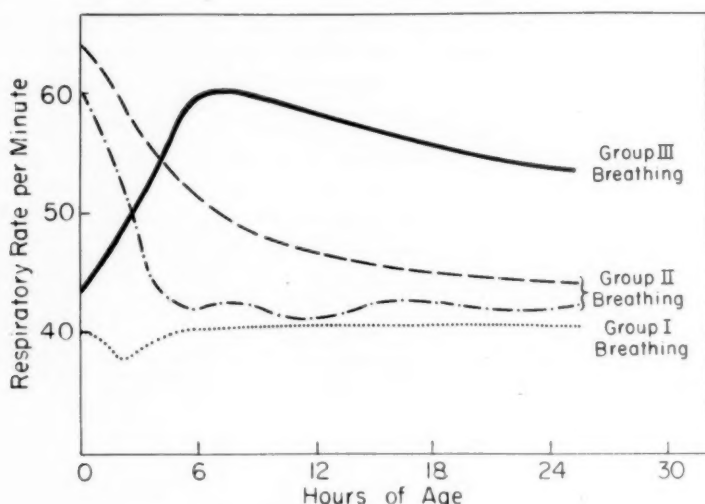


Fig. 1.—Breathing patterns in neonatal period.

Nevertheless, it can be observed that there were no deaths in premature babies, no matter how great the degree of prematurity, if the breathing pattern was normal.† With Group III breathing, on the other hand, there was a considerable mortality rate. This rose from 7 per cent in the slightly premature group to 36 per cent in the 1,000 to 1,495 gram group in the present series.‡

It has therefore seemed to us that to pursue the cause of death further, particularly in prematures, we must try to learn the cause of Group III breathing. Quite obviously, such a study could not be conducted until a sufficiently

*There were 2 deaths in the cesarean section group (neither classifiable as to breathing type). One was a baby with hydrops (nonerythroblastotic) who died in a few minutes and the other (also dead in about one hour) was delivered of a mother who was markedly hypotensive for a considerable period, after a very ineptly administered anesthetic.

†One baby in the 1,500 to 1,995 gram group died (after a normal hospital stay) at the end of two weeks. No autopsy; cause unknown.

‡Mortality figures throughout this study are exclusive of gross developmental anomalies although we do not really approve of "excluding" anything. We felt that these infants would have died regardless of how they breathed and in some groups their inclusion altered the percentages appreciably and was therefore confusing.

large series could be accumulated. It was hoped that the present series of 612 premature deliveries and 189 cesarean sections, with 216 full-term vaginal deliveries as controls, would be adequate. As stated above, this number has not proved large enough to be conclusive and our study is therefore being continued. This, then, is only a preliminary report.

TABLE II. MORTALITY WITH ABNORMAL BREATHING

	GROUPS I AND II		GROUP III	
	NO.	% MORTALITY	NO.	% MORTALITY
<i>Term Infants.</i> —				
Vaginal delivery	205	0	11	0
Cesarean section	138	0	17	0
<i>Prematures.</i> —				
2,000-2,495 grams	356	0	60	7
1,500-1,995 grams	80	0*	64	23
1,000-1,495 grams	12	0	39	36
Total	448	0	163	20

*Infant died in 2 weeks—cause unknown.

We have tried to investigate all possible angles with which abnormal breathing could be correlated. Age of the mother was first studied with the thought that abnormal breathing might be somewhat more frequent in the infants of older mothers. Such was not the case; perhaps it may prove to be so in a larger series. Similar studies of parity, prepregnancy body weight, weight gain in pregnancy, length of first stage of labor, and length and difficulty of second stage of labor yielded no correlation of significance. Markedly premature rupture of the membranes, more than 24 hours before onset of labor, did lead to more frequent abnormal breathing in this series. The difference (36 per cent versus 22 per cent in the whole premature group) would seem to be significant. Sixty patients with mild toxemia, 11 with severe toxemia, and 3 with eclampsia had roughly the same proportion of Group III babies as the nontoxemia patients. Forceps delivery had no effect. Breech-delivered babies behaved the same as those with cephalic presentations. There was a definite suggestion that the babies whose mothers had had little or no prenatal care did not do as well as those whose mothers had had good prenatal care, but, in the limited number available, this difference was not great. Analgesia, in the first part of the study, was principally interrupted nitrous oxide oxygen (25 per cent) and, later, combined interosacral⁵ and pudendal block with 1 per cent Cyclaine.* Drugs (usually 50 mg. of Demerol) were employed in 119 patients. No barbiturates were used. Anesthesia, beyond the regional analgesia previously administered, was, when necessary, also in the form of nitrous oxide in whatever increased percentages were needed. Almost all of the perineal stages could be conducted with no more than a 50 per cent mixture. In view of the recent report by Phillips,⁶ we expected that those babies delivered under regional analgesia and anesthesia alone would do better, but such was not the case. We have recently changed over to almost routine regional anesthesia, particularly for the premature babies, and must therefore look to other hospitals for comparative results with other forms of analgesia and anesthesia. An effort is presently being made in this direction.

Fortunately, our charts included records of mild and moderate degrees of delayed respiration and cry. Thirty seconds to 2 minutes was labeled as mild asphyxia even though no cyanosis or muscle flaccidity was present. From

*Cyclaine—hexylcaine hydrobromide—Merck Sharp & Dohme, Philadelphia.

2 to 5 minutes was labeled as moderate asphyxia and anything over 5 minutes was called severe asphyxia. Under this definition, mild asphyxia did seem to result in a somewhat higher percentage of Group III breathing later, but the contrast was not a sharp one. Moderate and severe asphyxia, on the other hand, seemed to show a marked increase in abnormal breathing (Table III) and subsequent mortality (Table IV). The number of patients involved is so small that it involves no more than a strong suggestion. The uniformity of the trends does somewhat strengthen the suggestion.

TABLE III. ASPHYXIA AND INCIDENCE OF ABNORMAL BREATHING

	NO ASPHYXIA			MODERATE AND SEVERE ASPHYXIA		
	TOTAL	GROUP III		TOTAL	GROUP III	
		NO.	%		NO.	%
Term infants	205	9	4	3	2	67
Prematures						
2,000-2,495 grams	392	55	14	8	3	38
1,500-1,995 grams	123	54	44	7	5	71
1,000-1,495 grams	40	28	70	6	6	100
Total prematures	555	137	25	21	14	67

TABLE IV. MORTALITY AFTER ASPHYXIA

WEIGHT (GRAMS)	GROUP III NO ASPHYXIA			GROUP III MODERATE AND SEVERE ASPHYXIA		
	NO.	DEATHS	%	NO.	DEATHS	%
2,000-2,495	55	3	5	3	1	33
1,500-1,995	54	9	17	5	4	80
1,000-1,495	28	10	36	6	3	50
Total prematures	137	22	16	14	8	57

One might well ask whether the same factors which produce delay in initial respiration and cry do not also operate to bring about Group III breathing later. It is, however, obvious from the tables that each can occur without the other. It is also apparent that the two, in combination, are much more serious than either alone. We hope to report a much larger series as soon as it can be compiled.

Summary

1. An abnormal form of breathing which serves to identify infants in potential difficulties has been re-emphasized.
2. All deaths in the series occurred in infants with this type of breathing.
3. Lack of prenatal care and a lag in the onset of labor, following premature rupture of the membranes, seem to contribute to the incidence of this abnormal breathing.
4. Analgesia and anesthesia, as employed in this series, did not affect the infants' breathing.
5. Asphyxia, even in moderate form, seemed to affect both the incidence of abnormal breathing and the subsequent mortality rate.

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Discussion

DR. ANTHONY D'ESOPPO, New York, N. Y.—The newborn have become so much an interest of the pediatrician that the scientific contributions related to this field of medicine appear with decreasing frequency in the literature devoted to our specialty. This is largely inevitable. When we gave over the care of the newborn to the pediatrician it was obvious and proper that he would also take on the research activities related to them.

However, our interest must continue. If our specialty deals with the totality of human reproduction it should include a concern about the newborn. This might express itself in some sort of teamwork between obstetrician and pediatrician such as the present contribution demonstrates.

The authors should be congratulated for their leadership in directing our attention to the respiratory patterns of the newborn, but they must also be prepared to believe that many of us have seen exceptions to the rules.

Respiratory rates of premature babies are not easily obtained. Breathing is frequently periodic and it is difficult to know whether to count only regular respirations or to include the periods of apnea.

Some very sick infants are born with slow, labored breathing which remains slow until death; some have high rates at birth which remain high. We must also recognize that the respiratory rates of the infant are influenced by a number of factors other than its internal milieu. Body position, environmental oxygen concentration, and environmental temperature may significantly affect the rate.

A problem of practical importance is that the classification of breathing types on a rate basis becomes prognostically useful only when the rate has been plotted from birth. For example, when the pediatrician sees a baby that is several hours old with a rate of 50, classification is difficult. It may be going in either the right or wrong direction.

Referring to the usefulness of the respiratory rate as a means of alerting us, it might be more practical to establish a recovery nursery where all newborn babies could be carefully observed for the first 24 hours of life. I am certain that this is the way our pediatricians would manage this important aspect of newborn care if they were given the opportunity to redesign our nurseries.

It is of interest to note the high correlation between asphyxia at birth and Group III breathing. This stresses the need for prompt and efficient resuscitative measures.

That there would be a lack of correlation noted between pregnancy and labor factors and the distressed type of breathing is understandable. Although pathologic data have not been mentioned in this paper, these probably include many diverse conditions. Hyaline membrane disease, congenital pneumonia, pulmonary hemorrhage, atelectasis, and congenital lesions of the heart and great vessels with cardiac failure are probably all represented in the autopsy material. We should therefore expect the etiology of Group III breathing to be multifactorial. Perhaps more significant correlations could be obtained if the clinical data, referable to the mothers of these babies, were related to the several categories of pathologic findings.

Dr. Calkins has shown that there is a twofold incidence of Type III breathing in babies delivered by cesarean section. He made the assumption that, on the basis of these data, the chances of fetal death following cesarean section would be at least double that following vaginal delivery! I am unable to understand why we repeatedly see this kind of unqualified reference to the increased fetal risk of the cesarean section without mention of the reasons for which the operations were done. Our own experience in nearly 2,000 healthy mothers subjected to repeat elective cesarean sections at term does not support this

view. As for the premature baby, the risk of the operation per se completely defies calculation because cesarean sections are not done in these cases unless there are complications which in themselves practically always pose distinct threats to the child.

In addition to our perinatal mortality data in cesarean sections, we have other material such as the Apgar scores, the time of sustained respirations, and biochemical indices which do not support the view that there is an increased risk to the cesarean section when the operative procedure is the sole insult to the child. We have seen a few cases in which an episode of maternal hypotension either from the spinal anesthesia itself or possibly from compression of the mother's inferior vena cava by the heavy gravid uterus while she remained in a prolonged supine position, may have been a factor in an asphyxiated infant with corresponding changes in oxygen saturation, blood pH, PCO_2 , and buffer base. These cases, in our experience, fortunately occur quite infrequently, and they could hardly significantly affect the perinatal mortality rate.

I may seem to belabor one of the small points in this paper that was quickly passed over in the text but I want to make certain that those who will read it do not come away with the misconception that the data presented here have in any way shown that there may be a special risk to the baby inherent in the cesarean operation itself. This is a most important consideration when, in the face of complications, one must weigh as best he can the many variables that lead him to choose between abdominal and vaginal delivery. In evaluating this question we must not confuse what are purely obstetric hazards with surgical risks.

DR. BENJAMIN TENNEY, JR., Boston, Mass.—This interesting presentation by Dr. Calkins has brought to our attention the importance of an abnormal breathing pattern in the newborn infant. There can be no question of the validity of this observation. As stated by the author, it should alert one to special care and observation of these abnormal infants. One should be most cautious concerning the prognosis of such a baby in dealing with the parents. The outcome cannot be predicted for at least 72 hours. Hyaline membrane disease is the common cause of death. Unfortunately, while an abnormal breathing pattern is an accurate prognostic sign, there is nothing that we can do to treat this disease.

It is of interest that premature rupture of the membranes does contribute to the incidence of abnormal breathing. If this could be shown to be due to the presence of infection of the newborn, this would be a prognostic sign of value in instituting treatment.

One would expect asphyxia to contribute strongly to this group of babies. This puts further emphasis on the danger of anoxia during the period of birth.

The increased incidence of Group III breathing in babies delivered by cesarean section, particularly in the premature, is further evidence that cesarean section is not the method of choice as far as delivery of the infant is concerned.

At our hospital, Dr. Sidney Gellis has found a marked increase in abnormal breathing pattern in many babies born of diabetic mothers. These infants may die in 24 to 72 hours from respiratory failure due to hyaline membrane disease. Some may improve and recover even with hyaline membrane. He believes that increased respiratory rate is a valuable prognostic sign.

In a comparable series, Dr. Gellis has found that the respiratory rates of babies born of diabetic mothers average considerably higher than the respiratory rates of premature infants born of normal mothers. This was true whether babies of diabetic mothers were born by cesarean section or through the birth canal. The respiratory rates of normal prematures were higher than those of full-term babies born by cesarean section. Term newborns delivered by cesarean section showed a higher respiratory rate than term newborns delivered by the pelvic route.

DR. EDWARD L. KING, New Orleans, La.—I want to ask several questions: Is this pattern varied according to the type of delivery? Can it be related to the type of sedation? Can it be employed as an aid as to what is the best type of sedation which will cause the least damage to the baby?

DR. CALKINS (Closing).—Dr. D'Esopo contended that cesarean section was no more dangerous to the baby than was vaginal delivery. I know of only one large series of cases, and I hoped that Dr. Davis would tell us about that. If I do not quote the Chicago statistics correctly, I hope he will say so. As I recall it, they have reviewed a very large series of cases where there were no complications in the mother; the cesarean sections were all repeat procedures or otherwise entirely normal ones. The neonatal death rate in this group was 0.9 per cent, as compared to 0.4 per cent of babies delivered vaginally. Is that correct?

DR. M. EDWARD DAVIS, Chicago, Ill.—If my memory serves me correctly, that is right.

DR. CALKINS: These were revealing figures to me. I think we must face the fact that cesarean section is not as safe for the baby as vaginal delivery, other things being equal.

Dr. King, I did not make it very clear in my presentation, but we did not find any correlation between the type of vaginal delivery, whether operative or spontaneous, and the type of breathing. There was one death following midforceps delivery, but this was immediate and the baby's breathing could not be typed so it did not appear in the figures. Similarly, our data with respect to analgesia and anesthesia were not revealing. We had fully expected that the results with regional anesthesia would be better than with general, but such was not the case and we can only say that this was probably because the amount of general anesthesia was not too great. We are now in the process of collecting data from other hospitals where more analgesia and deeper anesthesia are given, and I hope to have figures to report later.

THE MOVEMENT OF TRITIUM-LABELED WATER IN THE HUMAN OVARIAN FOLLICLE*†

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THE morphologic, chemical, and endocrinologic aspects of nonneoplastic cystic structures in the ovary have been extensively studied and well documented. Their dynamic aspects, however, have received less attention. By what mechanism and at what rate does fluid collect in such structures? What is the permeability of the barrier between the follicular liquor and the blood to water and dissolved substances? The answers to these and other questions have remained a matter of conjecture, often based on rather scant evidence.

In the past, secretion has been a process often suggested to explain the accumulation of water and dissolved materials in the Graafian follicle.^{1, 2} The presence of cells lining such cystic cavities has always suggested to the microscopist the possibility of secretory function and segregation of the follicular contents. The known growth response of the follicle to gonadotrophic hormones and follicular production of estrogens, as demonstrated by the presence of hormones in follicular fluid and the fluid from cystic corpora lutea,^{3, 4, 5} has been taken by some as evidence of secretion by the lining granulosa cells. More recently, radioactive sulfate has been shown to be incorporated into follicular fluid⁶ and is believed to be directly secreted into this fluid after accumulation in the granulosa cells.⁷

Simple *transudation* has also been suggested as a possible mechanism for the accumulation of fluid in nonneoplastic ovarian cystic structures.^{8-11, 15-18} Spiral arteries have been described in the ovaries of many species including man.^{8, 12-22} Reynolds and co-workers¹⁵⁻¹⁸ have proposed that these arteries may act as an arterial pressure regulating mechanism for the ovary. Presumably, disturbances in this pressure mechanism, such as straightening or irregular uncoiling of the spiral vasculature, could cause excessive hydrostatic pressure, creating net movement of fluid into the follicle by filtration against a lower internal pressure. Burr and Davies²³ have observed a marked increase in vascularity and actual swelling of the ovaries in rabbits just prior to ovulation. Clamping of the ovarian vessels or obstruction of the venous return at

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this time caused doubling of follicular size and, in some instances, rupture in a matter of seconds. Dr. John Clark as long ago as 1900⁸ described similarly striking results during injection studies on an excised human ovary.

Zachariae and Jensen²⁴ have demonstrated differences in *osmotic pressure*, viscosity, and elastic recoil in bovine follicular fluid from follicles at different stages of development. They suggest that these changes result from enzymatic splitting of mucopolysaccharides with resulting net transfer of fluid into the follicle, possibly leading to ovulation.

Such concepts suggest a high degree of permeability of the barrier between the blood and follicular antrum. Experiments with trypan blue²³ and sky blue²⁵ have demonstrated a considerable permeability of the blood-liquor barrier in rabbits under certain conditions but no similar observations have been made on man. The experiments to be described were designed to gain information regarding the permeability to water of the blood-liquor barrier in the human female.

During the course of these experiments von Kaulla and co-workers²⁶ reported that I^{131} and other labeled molecules of greater size appear in human follicular fluid within a relatively short time after intravenous injection (as little as 15 minutes for I^{131}). These observations lend support to the conclusion reached in this study that the water in follicular fluid is by no means isolated from the water in the circulating blood plasma, the blood-liquor barrier being highly permeable to these molecules.

Material and Methods

Studies have been carried out upon women in the menstrual age group operated upon for uterine pathology. Between 1.5 and 5 mc. of tritiated water was administered intravenously at various times preoperatively and during the course of operation. Heparinized venous blood samples were obtained at appropriate intervals and the plasma separated by centrifugation for counting. The follicular fluid was obtained at laparotomy through a 25 gauge needle. After experimentation it was found that the needle could be passed obliquely through the wall of the follicle at the most avascular spot often with little or no leakage of fluid. Generally, single samples were free of blood. Grossly bloody samples were discarded. On one occasion three samples were drawn from a single follicle (Patient M. W.). A tiny pellet of blood cells was observed in the second and third samples after they were centrifuged. This degree of contamination in a total estimated volume of 4 ml. is not believed likely to be significant. Samples were counted in a Packard Tri-Carb Model 314-DC Liquid Scintillation Spectrometer, the background varying between 68 and 88 disintegrations per minute (DPM). The method used was essentially the same as that recently reported by Chen.²⁷ One hundred gamma of follicular fluid or plasma was complexed by the addition of 1.0 ml. of the quaternary base Hyamine and dissolved in phosphor-containing toluene with the addition of ethancl. Correction for quenching was made by recounting after the addition of an internal standard to each sample. Histologic examination of the follicles sampled was attempted when the ovary was removed. The results were quite unsatisfactory because of hemorrhage and disruption of the follicular contents. Therefore, we are unable to say which, if any, of these follicles were undergoing atresia. Grossly, they all appeared healthy, the largest being 2 cm. in diameter and the smallest about 0.5 cm. in diameter.

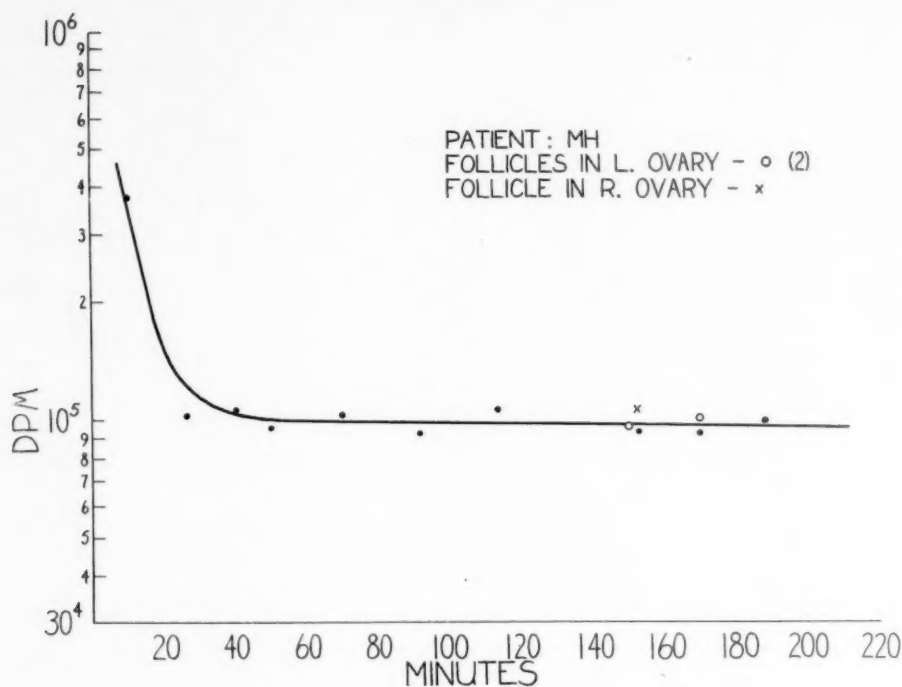


Fig. 1.

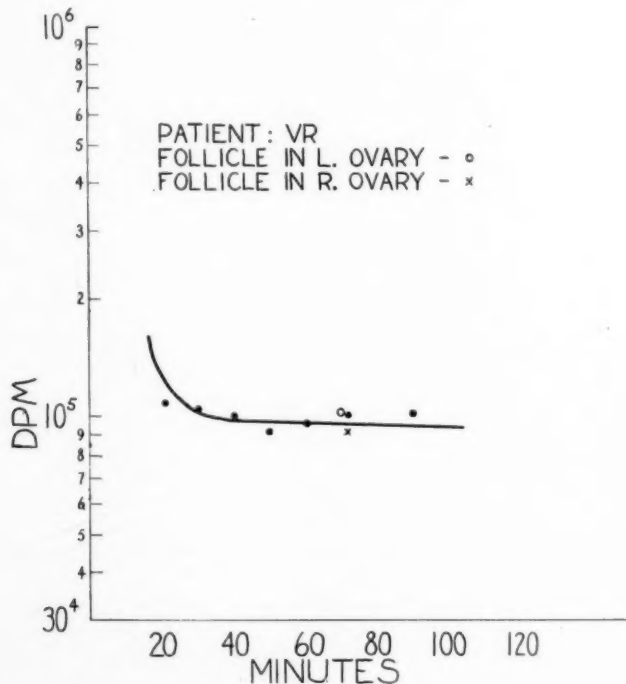


Fig. 2.

Figs. 1-3.—Peripheral venous dilution curve and follicular concentrations for tritiated water after intravenous injection (in disintegrations per minute). The follicular samples, which have equilibrated with the blood, are all single samples.

Results

These data are presented in Figs. 1-4. The curves represent the decreasing venous concentrations of tritium after intravenous administration. Rapid equilibration with body water occurs, being complete in most instances at the end of one hour. These curves are seen to vary considerably from patient to patient in the first few minutes, a phenomenon which has been noted after the intravenous administration of deuterium. These differences are probably best explained by the variable rates of tissue equilibrium involved.²⁸ In some patients fluctuations in peripheral venous concentrations have been noted even after equilibration. We are inclined to believe that such fluctuations result from the administration of intravenous fluid at varying rates.

It will be noted that the follicular fluid obtained from Patient M. H. had equilibrated with the blood. For this reason samples from subsequent patients were obtained at progressively earlier intervals after administration of

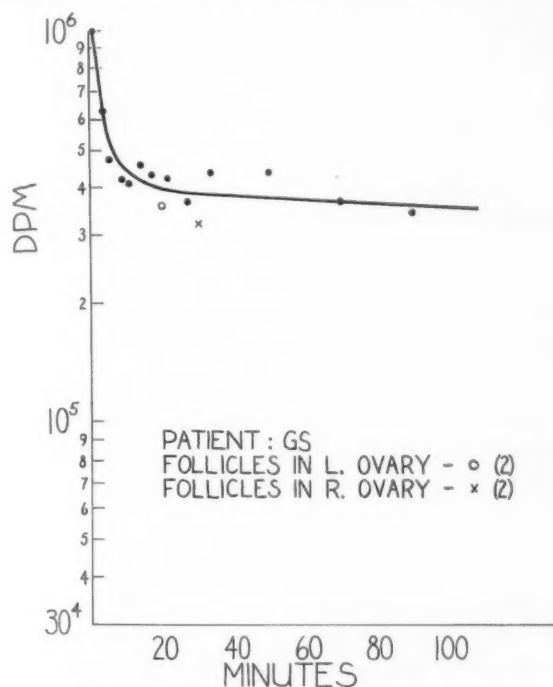


Fig. 3.—For legend see opposite page.

the tritiated water. It was found that equilibration had occurred prior to 70 minutes (Fig. 2) and even prior to 30 minutes (Fig. 3). Because of the problems occasioned by the mixing and variable tissue equilibration rates of tritiated water in the *peripheral* blood, venous samples from the next patient were obtained from an ovarian vein immediately adjacent to the follicle.

At operation for preinvasive carcinoma of the cervix it was ascertained that a follicle 2 cm. in diameter (about 4 ml. volume) was present in one ovary and a needle was placed in one of the adjacent ovarian veins. The tritiated water was administered via a vein in the foot and samples of venous blood were obtained at 2, 3, 4, 7, 9, and 12 minute intervals. Simultaneously, samples of follicular fluid were obtained at 4, 7, and 9 minutes after administration. It is apparent (Fig. 4) that the tritium concentration rises extremely rapidly in the follicular fluid.

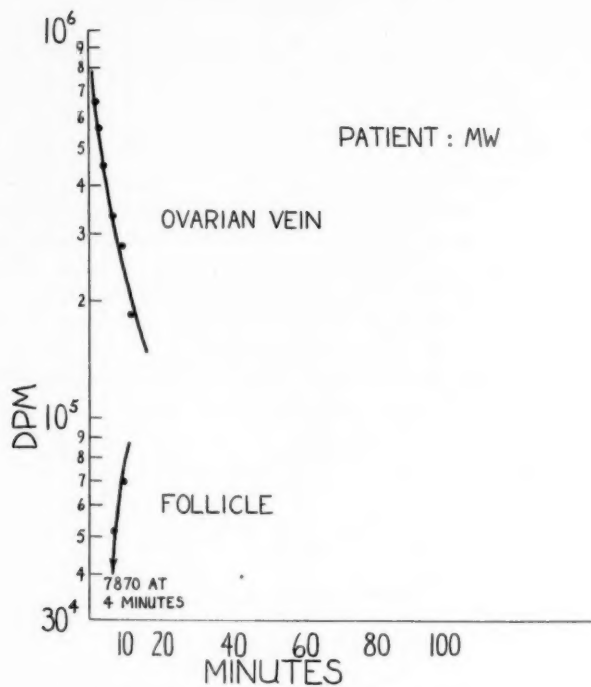


Fig. 4.—Concentration of tritiated water in samples drawn simultaneously between the second and the twelfth minute after intravenous administration from an ovarian vein and a single follicle, 2 cm. in diameter (see text).

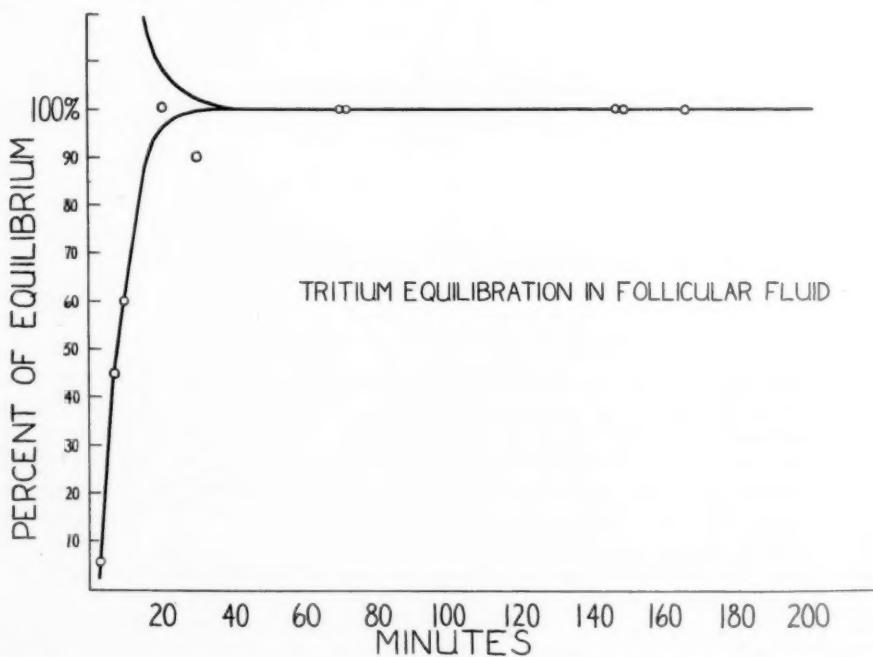


Fig. 5.—The combined data, each point calculated as the percentage of the equilibration value for each patient, are plotted against time. The venous dilution curve (upper curve) is a purely empiric one.

These data have been combined and are presented as the per cent of the equilibration value for each patient. A theoretical venous dilution curve (empirie) has been constructed for comparison (Fig. 5).

We estimate that equilibration generally occurs in the ovarian follicle at some time between 15 and 25 minutes after intravenous administration of tritiated water and well before general body equilibration has occurred.

Comment

The validity of all tracer studies hinges on the basic assumption that labeled molecules follow their unlabeled counterparts faithfully. There is evidence that, in studies of this type, tritium-labeled water does this without significant isotope effect.³¹ Granting this basic assumption, it seems apparent that there is very rapid exchange of water molecules between the vascular compartment and the follicular antrum. The fact that there is no observable change in follicular volume during the course of the experiment suggests that this exchange is by the mechanism of diffusion *without net transfer of fluids*. The speed with which equilibration occurs suggests further that these diffusion rates approach those of the capillary. It is entirely likely that the rate of exchange is limited by the vascularity of the region or structure rather than by any "membrane" effect. Such limitation by vascular supply has been demonstrated for heart and gastrocnemius muscle by the deuterium studies of Johnson and associates²⁹ and for skeletal muscle of the hind limb by Pappenheimer.³⁰

As pointed out, these data do not bear directly on the problem of net transfer. It seems obvious, however, that the blood-liquor barrier is highly permeable to water molecules and, therefore, would not seem likely to interfere seriously with passive transfer of fluid due to combined hydrostatic and osmotic forces.

Summary

To determine the permeability of the barrier between the follicular fluid and the vascular compartment to water, tritium-labeled water has been used as a tracer. Rapid appearance of the tracer within the follicular antrum suggests that this barrier is highly permeable to such molecules and that rapid exchange of water does occur by diffusion. The implications of this observation are discussed.

All samples were counted by Mrs. Lynn Gilboe under the direction of Dr. Charles Heidelberger in the McArdle Cancer Research Laboratories. We are most grateful to Dr. Heidelberger and his staff for making these studies possible.

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Discussion

DR. SOMERS H. STURGIS, Boston, Mass.—The development of the ovarian follicle from a small structure containing an egg and one or two layers of granulosa cells and an antrum to the fully organized, large, cystic follicle just prior to ovulation is signalized by a number of features that are not at all understood at the present time. Chief among these may be mentioned the accumulation of fluid. Is this a transudate or a secretion from the granulosa cells of the ovary? Is the accumulation of follicular fluid attended by increased pressure relationships that provide a mechanism, as it were, for squeezing the maturing follicle outward to the cortex of the ovary? The authors of this paper have addressed themselves to some of the basic mechanisms that may answer these unknowns. They have reported the first observations on the exchange of labeled water from the general circulation into the follicle across what is termed the blood-liquor barrier.

I would like to raise a voice in objection to the concept of a barrier. This is a concept borrowed from the widely accepted use of the term "barrier" to clarify passage of electrolytes, steroids, and other compounds from maternal circulation across the cellular membrane in the placenta that separates these substances from the fetal circulation. I think it is inappropriate, however, to apply this concept to an intrinsic part of the ovary itself, such as the growing follicle, with the intimation that there is anything that separates it from its environment. The data in the 4 cases of normal follicles lend support to the conclusion that equilibration of tritium-tagged water into the follicular liquor takes place rapidly, in a matter of 20 to 30 minutes. When the authors apply their technique to a study of pathologic ovarian cysts, the problem of interpreting the data becomes vastly more complex. Intravenous fluids were being given simultaneously at different rates. In one tumor, three different locules with a different quantity of fluid in each were tapped. Thus the pressure relationships and volumes of each were varied, and equilibration into and concentration of labeled water in each must have been affected by the demands of the technique. The authors have rightly pointed out these pitfalls, especially in relation to large structures, as in one case of hydrosalpinx.

There are very few other pertinent reports of similar work with which to compare these observations. The authors note that I^{131} is found in follicular fluid in 15 minutes and Morigard has found S^{35} in ovarian follicles, also shortly after injection into the general circulation. The conclusion that a rapid exchange of labeled water molecules

occurs, probably by diffusion into the follicular liquid, appears valid. This does not, of course, answer as yet the problem of the origin of the rapid increase in liquor folliculi during the last few days of rapid increase in size of the maturing follicle. The answer to some of the basic mechanisms involved in maturation of the ovarian follicle will have to depend ultimately on the kind of basic work represented by these studies of Drs. Peckham and Kiekhofers.

DR. PECKHAM (Closing).—The term "barrier" does not originate with me. This term has been used by many authors interested in the ovarian follicle, most recently F. Zachariae. While semantically this term may not be ideal, it has been used here for want of a better term. I will happily accept suggestions in this regard.

THE EFFECT OF TESTOSTERONE ON EXPERIMENTAL ENDOMETRIOSIS IN RHESUS MONKEYS*†

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EXTERNAL endometriosis as a benign disease is limited in its growth to the years of menstrual function; therefore, the implication that this growth is closely related to the circulating estrogens and progesterone is a natural one. Androgens are antagonistic to estrogens and, in addition, they depress pituitary function. For these reasons testosterone has been recommended in the treatment of clinical external endometriosis. The effect of testosterone on the gross and microscopic appearance of the lesions has not been adequately studied and reports of beneficial effects have been essentially clinical impressions based on symptomatic relief.

External endometriosis can be produced in rhesus monkeys by surgical transplantation of endometrium. The areas of endometriosis can then be inspected grossly and subjected to biopsy periodically during the course of any given study. By this method the gross and histologic effects of long-term testosterone administration upon the endometriosis could be evaluated in these animals. The authors have studied and reported the effects of diethylstilbestrol, estrone, and estrone and progesterone on experimentally produced endometriosis in monkeys.^{11, 12}

Material and Methods

Eight adult female rhesus (*Macaca mulatta*) monkeys were used. One animal had extensive pelvic endometriosis which had been found 2 years after a hysterotomy for a near-term pregnancy. Five of the animals had been used in other experiments which did not alter an essentially normal ovarian and uterine function. Three monkeys had large fragments of endometrium transplanted into the anterior abdominal wall beneath the rectus muscle, and in 4 other animals additional endometrial transplants were made into the uterine wall and one ovary. Practically all of the transplants of endometrium remained viable in the new locations and a base line of gross and microscopic appearance was established for each animal before the administration of testosterone.

*Presented at the Eighty-second Annual Meeting of the American Gynecological Society, Hot Springs, Va., May 21-23, 1959.

†This investigation was supported by Research Grant C-2892 from the National Cancer Institute, U.S. Public Health Service. The testosterone was provided by Ayerst Laboratories, New York, through the courtesy of the Medical Director, Dr. John B. Jewell.

Twice weekly an intramuscular injection of 0.25 c.c. of an aqueous suspension of pure crystalline testosterone (1 c.c. equals 25 mg.) was administered to each animal. Thus, each animal received approximately 6 mg. of testosterone twice a week; the aqueous medium permitted slower absorption than an oil medium would have. The testosterone was administered on this schedule for a period ranging from 6 to 27 months. Periodically during this time and terminally the areas of ectopic endometrium were surgically investigated and biopsied.

Results

Monkey 839.—This animal was received at the Carnegie Institution of Washington, Department of Embryology, directly from the importer on Oct. 4, 1944. It was used for some experimental studies with Prostigmin and progesterone. On May 13, 1954, a 155 day gestation was removed by hysterotomy. At laparotomy on May 8, 1956, a 4 cm. cystic mass filled with chocolate-colored material was found in the pelvis, almost encasing the pelvic organs and agglutinated to the bowel and the bladder. The wall was thick and the inner surface had a reddened membrane, in places almost papillary. Microscopically, the cyst was endometrial in type with nonsecretory endometrium, and there were numerous pigment-laden macrophages in the wall. Testosterone was begun on Sept. 24, 1956, and 14 and 27 months later the gross and microscopic findings were essentially identical to those noted before treatment (Fig. 1).



Fig. 1.—Endometrial tissue lining cyst in Monkey 839 after 14 months of testosterone injections. ($\times 150$; reduced $\frac{3}{4}$.)

Monkey 897.—This animal was received from the importer on Jan. 4, 1949. It was used in attempts to perform Rubin's tests. Subsequently an abdominal hysterotomy was done (Dec. 18, 1951) and all of the endometrium was surgically transplanted beneath the rectus fascia into the muscle. The transplant grew actively to a 2.8 cm. nodule of endometriosis. A biopsy of this nodule before testosterone treatment was begun showed healthy endometrial tissue as well as an interesting accumulation of endometrial cells in an endothelium-lined channel (Figs. 2 and 3). Testosterone was begun on Dec. 27, 1954. The gross dimension of the abdominal wall nodule did not change appreciably. Biopsies 5 months and 10 months later, as well as autopsy studies 17 months after the beginning of testosterone administration, revealed active endometriosis with nonsecretory

endometrium. Although the animal had only 3 menstrual periods after beginning testosterone, the biopsy after 5 months showed evidence of old and recent hemorrhage within the endometriosis (Fig. 4). Death was due to tuberculosis.

Monkeys 959, 961, 962, 964, 965, and 966.—Four of these animals had been used in a surgical experiment consisting of shifting the vagina into a gluteal muscle. These experiments were unsuccessful because of a rapid tissue breakdown and the formation of vaginoperineal fistulas. In 2 animals endometrium

Fig. 2.

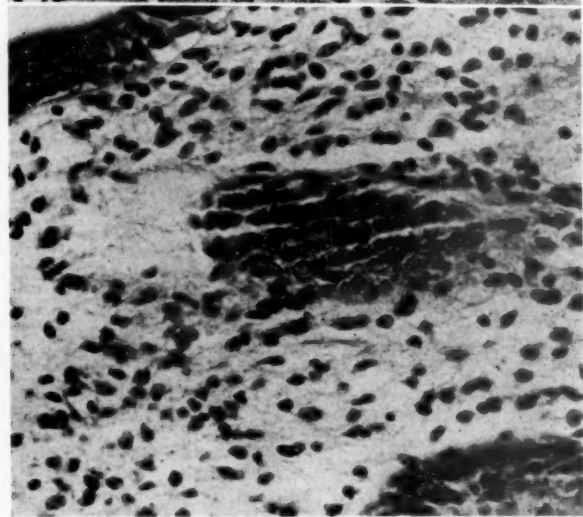
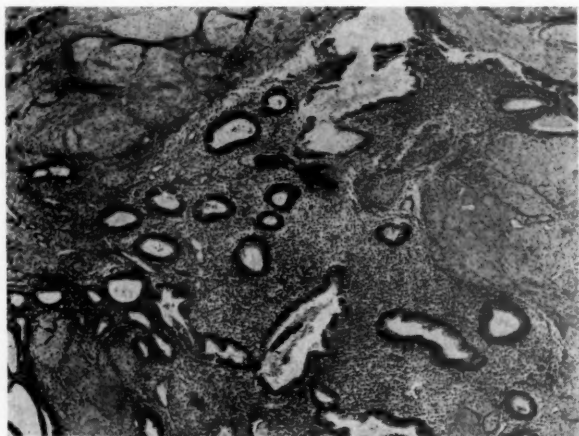


Fig. 3.

Fig. 2.—Biopsy of anterior abdominal wall endometriosis in Monkey 897 prior to initiation of testosterone treatment. Arrow marks endothelium-lined channel containing endometrial epithelium. (See Fig. 3 for higher power magnification of the marked area.) ($\times 50$; reduced $\frac{1}{4}$.)

Fig. 3.—Higher power of area marked by arrow in Fig. 2. This appears to be an endothelium-lined channel surrounded by endometrial stroma and containing endometrial epithelium. ($\times 500$; reduced $\frac{1}{4}$.)

was surgically transplanted to the anterior abdominal wall and in the other 4 animals the transplants were into the uterine wall, one ovary, and the abdominal wall. Biopsy of most of the transplants was carried out after several months and they were found to be viable endometriosis. Testosterone was then begun.

The transplants were periodically biopsied over a 6 to 26 month period: No. 959—9 months; No. 961—11 months; No. 962—16 months; No. 964—26 months; No. 965—17 months; and No. 966—6 months. Practically all areas of endometriosis continued to show healthy endometrium without evidence of excessive fibrosis and certainly no atrophy. It was particularly interesting that the abdominal wall transplant in Monkey No. 962, after 13 months of testosterone administration, contained many endometrial glands of a late secretory phase.

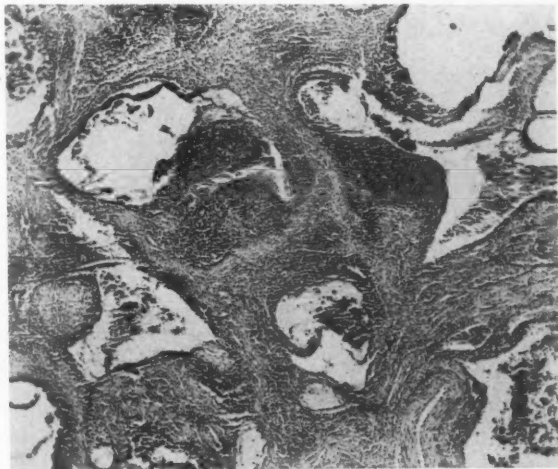


Fig. 4.—Anterior abdominal wall endometriosis in Monkey 897 after 5 months of testosterone therapy. Note the evidences of old and recent hemorrhage. ($\times 50$; reduced $\frac{1}{4}$.)

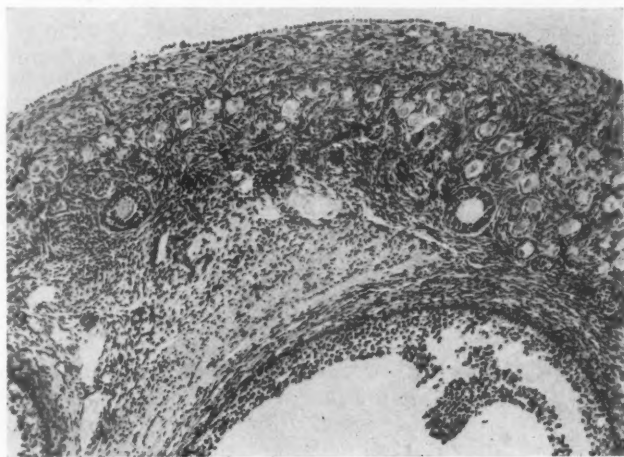


Fig. 5.—Cortex of ovary of an untreated adult female rhesus monkey. ($\times 100$; reduced $\frac{1}{4}$.)

Summary of Results.—Eight rhesus monkeys with endometriosis were used to test the influence of twice weekly injections of testosterone, 6 mg. each, upon the ectopic endometrium. One animal had extensive pelvic endometriosis, including an endometrial cyst which followed a hysterotomy 2 years before, and the other animals had endometriosis produced by surgical transplantation of normal endometrium into the abdominal wall and/or pelvic areas. Gross and microscopic studies of the endometriosis were made over a treatment period ranging from 6 to 27 months. All of the animals developed some signs of

masculinization, but, where it was possible to test the animals, vaginal bleeding occurred at irregular and infrequent intervals. Over this period of observation there was no evidence of atrophy or excessive fibrosis of the transplants; the ectopic endometrium remained healthy, usually showed areas of recent and/or old hemorrhage, and, except for one instance of a late secretory pattern, was non-secretory and proliferative in glandular pattern. In 6 animals an uninvolved ovary was studied histologically at the end of the treatment period; all demonstrated a variable degree of fibrosis of the tunica without underlying follicular cystic change (Figs. 5 and 6).

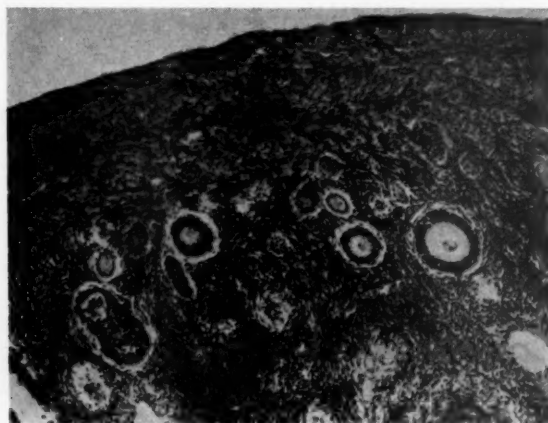


Fig. 6.—Cortex of ovary of Monkey 961 after 11 months of testosterone therapy. Note the increased fibrous thickening of the tunica as compared to Fig. 5. ($\times 120$; reduced $\frac{1}{4}$.)

Comment

The effects of various hormones and hormone combinations upon experimental endometriosis in monkeys have been reported by the authors. Four uncastrated rhesus monkeys were given daily injections of diethylstilbestrol in sesame oil over a period ranging from $3\frac{1}{2}$ to 24 months for a total dosage varying from 390 mg. to 36,560. There was no evidence of atrophy of the endometriosis; in fact, hyperplasia of the ectopic as well as of the normally located endometrium developed in the 2 animals under the longest treatment. In another series of experiments endometrial transplants were made and the animals castrated; one group received no hormones, another constant estrone, a third group constant estrone and constant progesterone, and a fourth group constant estrone and intermittent progesterone. Over an observation period up to 37 months the major local growth of the ectopic endometrium was found in the group receiving constant estrone and intermittent progesterone, i.e., in the group under a hormone schedule simulating the conditions of normal ovarian function. There was an interesting correlation between bleeding from the uterus and evidence of bleeding in the transplants. From these results we postulated that uninterrupted cycles of constant estrone and intermittent withdrawal of progesterone was the main stimulus for local growth and spread of endometriosis, provided the ectopic endometrium is not too fibrotically encased or cystically compressed to receive the full influence of the circulating hormones.

The administration of testosterone will inhibit menstruation in normal monkeys,^{5, 14} prevent estrogen withdrawal bleeding,^{1, 2, 5} and prevent bleeding following castration.⁵ The developments in the endometrium induced by estrogen in castrates could be maintained by testosterone without involution or

progestational changes and when bleeding was inhibited following injections of progesterone the progestational condition of the endometrium was preserved.² Hisaw⁸ found that testosterone in adequate dosage to castrated and immature female monkeys prevented estrogen withdrawal bleeding, precipitated bleeding in the presence of maintenance doses of estrogen, inhibited estrogen in enlargement of the cervix and metaplasia of the cervical glands, greatly decreased cornification of the vaginal mucosa, and brought about loss of edema of the sexual skin. In many respects, but not all, the effects of testosterone were similar to those of progesterone. Testosterone had about one-third as much effectiveness per milligram as testosterone propionate.

Wilson¹³ reported the first clinical use of testosterone for the therapy of external endometriosis. Since Hartman⁵ and Zuckerman¹⁴ found that the ovarian cycles in monkeys could be depressed by testosterone for the duration of treatment and a similar effect occurred in women,^{3,9} Wilson¹³ rationalized the use of this hormone for a 28-year-old patient with a symptomatic endometriosis nodule in the rectovaginal septum. The patient had refused operation or radiation castration. She received 4,800 mg. of testosterone over 13 months; the pelvic pain was relieved, there was a marked reduction in size of the endometriosis nodule, there were 5 episodes of scant vaginal bleeding, there was atrophy of the endometrium, but hoarseness, hirsutism, and enlargement of the clitoris developed. Normal menses resumed 5 weeks after discontinuation of treatment; the symptoms returned, and the nodule increased progressively in size. X-ray castration was then acceptable to the patient.

Hirst⁶ in 1943 was impressed with the symptomatic relief obtained from the use of testosterone in 2 patients with endometriosis. In 1947 he⁷ listed 19 patients treated with testosterone and noted the reduction in pain and tenderness, as well as the diminution in size of the endometrial cysts. Preston and Campbell¹⁰ in 1953 used oral methyl testosterone for 187 patients with clinical endometriosis (in only 16 instances was the diagnosis confirmed by subsequent operation). They did not exceed a monthly total of 300 mg. and the average course was 4 to 6 months. Dysmenorrhea and dyspareunia were completely relieved in about 80 per cent of the patients and the majority continued to have an arrest of symptoms for 6 to 12 months after testosterone was discontinued. They noticed a diminution in size of the lesions by two-thirds and a loss of tenderness to palpation. Forty-eight of the 80 patients with associated infertility became pregnant while on treatment, a remarkably high percentage. These results are reasonable evidence that the prescribed amount of methyl testosterone did not inhibit ovulation. Preston and Campbell¹⁰ recognized operation as the treatment of choice for patients with advanced endometriosis, but they did suggest this form of hormonal treatment for symptomatic relief during the earlier phases of the disease.

In the human it has been estimated that 500 mg. or more of testosterone a month will inhibit normal ovulation and menstruation. Testosterone propionate in comparatively large amounts (approximately 25 mg. twice weekly) will inhibit follicle growth and ovulation in the rhesus monkey¹⁴; under this treatment vaginal desquamation is reduced to practically zero, there are no pro gravid changes in the endometrium, but the clitoris enlarges markedly and the sex skin stays red without swelling. In our present experiments, with the use of approximately 6 mg. of an aqueous suspension of testosterone twice weekly, no accurate evaluation of the effect upon ovulation can be made. There were episodes of scant vaginal bleeding on occasions; one animal showed a recent corpus luteum in the ovary while under treatment, and another animal demonstrated a secretory pattern in the ectopic endometrium. Ovulation must have occurred on occasions, but growth of the clitoris was a prominent feature in all

animals, even in those with the shortest treatment—6 months. A normal ovary was studied in 6 of the 8 animals during treatment and in all of these there was some thickening and fibrosis of the tunica albuginea, but cystic follicles did not develop. This change in the tunica may simply be secondary to a degree of pituitary inhibition and slightly reduced ovarian activity. The Stein-Leventhal syndrome in the human is associated with a thickened ovarian tunica and frequently with hirsutism. Many writers on this subject have been impressed with the frequency of a high normal androgenic activity of the adrenals. The present experiments and the findings in the ovaries give a little support to the suspicions that a relatively increased androgenic influence plays a part in the Stein-Leventhal syndrome.

Summary

Androgens have been recommended in the treatment of external endometriosis on the basis of their antiestrogenic and pituitary inhibiting effects. Most reports have been based upon clinical impressions with little or no information about the gross and microscopic effects upon the ectopic endometrial lesions.

Eight female, noncastrated rhesus monkeys with surgically produced external endometriosis were given 6 mg. of an aqueous suspension of testosterone twice weekly for a period ranging from 6 to 27 months. By periodic observations and microscopic studies there was no evidence of atrophy or of fibrosis about the lesions. One biopsied lesion contained secretory endometrium, but all of the other biopsies had a nonsecretory, proliferative pattern with evidence of old or recent hemorrhage. In 6 of the 8 animals an uninvolved ovary was studied and all of these had a relatively thickened and fibrotic tunica; this finding may give some support to the theories of increased androgenic activity in the human disorder termed Stein-Leventhal syndrome.

The clinical reports of the use of androgens for external endometriosis are reviewed. From the clinical experiences this form of treatment may have some temporizing beneficial effects, but neither the clinical reports nor these experiments with rhesus monkeys warrant claims of atrophy or disappearance of the lesions.

We wish to express our appreciation to Dr. James D. Ebert, Director of the Department of Embryology of The Carnegie Institution of Washington, Baltimore, and his staff for their cooperation in this study and for the use of their facilities.

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Discussion

DR. GEORGE H. GARDNER, Chicago, Ill.—This is a report of a series of observations on the effects by testosterone on the gross and microscopic appearance of the experimentally produced endometriosis harbored by 8 adult female rhesus monkeys. The authors were of the opinion that such information was sorely needed since much that has been reported about the beneficial effects of testosterone on clinical external endometriosis in reality is nothing more than clinical impressions based solely on the symptomatic relief experienced by some patients. Furthermore, Drs. Scott and Wharton were particularly qualified to conduct experiments of this type since they had previously studied and reported the effects of diethylstilbestrol, estrone, and estrone and progesterone on such endometriosis in monkeys.

Accordingly, 8 of their monkeys with endometriosis were selected for this particular experiment. These were animals which had been under observation sufficiently long so that both the gross and microscopic characteristics of individual lesions in each animal had already been carefully observed and documented. Thereafter, each animal was given intramuscular injections of an aqueous suspension of 6 mg. of pure crystalline testosterone twice weekly, and these were continued regularly for varying periods of time—the shortest being 26 weeks and the longest, 27 months. Periodically, while the injections were being given, and also when they were concluded, the areas of endometriosis were surgically inspected and biopsy specimens were taken.

The effects of the testosterone were essentially the same in each of the 8 animals—all developed signs of masculinization; in those which could be tested, there was vaginal bleeding but this occurred irregularly and infrequently; none of the lesions showed either atrophy or notable fibrosis. In fact the endometriosis remained healthy and contained areas of hemorrhage, either recent or old; in all but one, the gland pattern was proliferative.

Hence, these experiments with rhesus monkeys fail to furnish any support whatever for the clinical claim that testosterone therapy will produce atrophy, let alone disappearance of the lesions of external endometriosis.

DR. SCOTT (Closing).—Clinically I have had occasion to use testosterone, particularly in those patients with recurrence of endometriosis with symptoms—patients in whom I know that the next surgical procedure to relieve them is going to have to be a complete one; so I use it in a palliative fashion. More recently I have been quite impressed with some close relatives of testosterone, the newer progesterone-like preparations, which have been worked on and studied thoroughly by Dr. Robert Kistner and the Andrews brothers and Dr. Strauss. I am pleased with the results from two of these preparations and now prefer them to testosterone.

URINARY ESTROGENS IN WOMEN*†

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BETWEEN the years 1929 and 1939, 3 estrogens, estrone, estriol, and estradiol 17β , were isolated and identified in human pregnancy urine. These 3 will be herein referred to as the "classical estrogens" since it was not until the years 1953 to 1958 that 7 more compounds structurally related to them were isolated and identified as natural products in the urine of pregnant women. (They are all referred to as estrogens, though not all of them exhibit estrogenic potency.) It seems more than likely, both from theoretical considerations and from the findings to be presented below, that others will shortly be added to the list.

A practical means of measuring urinary estrogens in women should yield information of clinical value in the diagnosis and treatment of patients. With so many estrogens to measure, the problem of methods, which has always been a stumbling block, becomes even more acute. Until 1955, bioassay was the only method of obtaining even reasonably satisfactory measurements, especially in the nonpregnant. In that year Dr. J. B. Brown of Edinburgh published a system of chemical assay¹ which has stood up well under a number of tests for precision, accuracy, and specificity in the measurement of as little as 2 to 4 μg of estradiol, estrone, or estriol per 24 hour volume.²⁻⁴ Through the courtesy of Dr. Brown, this laboratory was given the details of his method in August, 1954, and embarked upon a systematic investigation of the optimum conditions for the hydrolytic cleavage of estrogen conjugates as they occur in the urine of women, since there had never been agreement on this important point. In Brown's system of chemical assay, the method of acid hydrolysis proposed by Stevenson and Marrian⁵ in 1947 was adopted, with the realization that it was not ideal and that it resulted in variable losses of all 3 of the classical estrogens.⁶ Our findings in pregnancy urine⁷ led to the conclusion that enzymatic hydrolysis with bacterial glucuronidase, first proposed by Buehler, Katzman, and Doisy⁸ in 1949, yields considerably more of all 3 estrogens than is recovered by any single-step measure with acid and heat, but that mild acid hydrolysis of the residual urine, after extraction of the glucuronidase hydrolysate, is necessary

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for maximum recovery. In nonpregnancy urine this two-step procedure (glucuronidase plus acid) is particularly important since it yields 30 to 60 per cent more of each of the classical estrogens than is recovered after glucuronidase alone, after the hydrolytic measure used by Brown, or after any other single-step measure with heat and acid.⁹ Glucuronidase plus acid, therefore, has been adopted in this laboratory for the measurement of the classical estrogens. Analyses of results on the urine of nonpregnant women⁹ indicate that this change in the method of hydrolysis does not detract from the precision, sensitivity, and specificity of Brown's chemical assay for estradiol, estrone, and estriol, and considerably improves its accuracy, since the variable losses resulting from one hour of heat and acid do not pertain. The lack of uniformity of results when one hour with acid is used is shown in Table I.

Primarily as a check on the specificity of the chemical method, each extract that was chemically assayed in the above studies was also bioassayed. An offshoot of this part of the work has been the discovery that a number of compounds other than estradiol, estrone, and estriol are recovered in the phenolic fractions and often contribute as much to the estrogenic potency of the urine of

TABLE I. INFLUENCE OF METHODS OF HYDROLYSIS ON YIELDS OF ESTRIOL, ESTRONE, AND ESTRADIOL (CHEMICAL ASSAY)

METHOD OF HYDROLYSIS	PER CENT OF MAXIMUM* (AVERAGE \pm STANDARD DEVIATION)		
	ESTRIOL	ESTRONE	ESTRADIOL
<i>Pregnancy Urine (12 Specimens; 25 Weeks to Delivery).—</i>			
Glucuronidase alone	88 \pm 4 (2.3)†	83 \pm 4 (2.3)	79 \pm 8 (8.3)
One hour acid at 100° C.	68 \pm 10 (15)	65 \pm 13 (29)	65 \pm 15 (37)
<i>Nonpregnancy Urine (12 Specimens; Various Stages of Normal Cycles).—</i>			
Glucuronidase alone	67 \pm 9 (9)	63 \pm 9 (9)	65 \pm 13 (29)
One hour acid at 100° C.	61 \pm 18 (53)	64 \pm 18 (48)	78 \pm 17 (22)

*Maximum recovery of each, in all specimens, was after the two-step system of glucuronidase followed by 10 minute acid that was adopted. The results on glucuronidase alone and on one hour with acid (aliquots of the same specimens) are expressed as per cent of the maximum.

†The numbers in parentheses are the chi-square values, any figure above 8 denoting unsatisfactory uniformity.

both pregnant and nonpregnant women as do these 3; sometimes considerably more than they do. The yields and characteristics of these unknowns have been found to vary with the hydrolytic measure employed, and differences in their lability and solubility characteristics have indicated the presence of at least 6, 4 of which are common constituents of both pregnancy and nonpregnancy urine. The types of unknowns found and their rates of excretion have followed definite patterns in relation to known placental changes in pregnancy and to stages of ovarian activity in the nonpregnant. Pregnancy findings have already been published^{10, 11} and some of our results on chemical versus bioassay in nonpregnancy situations will be reported in detail elsewhere. It is the object of the present paper to present our evidence for the presence and physiological significance of these unknown constituents in the urine of both pregnant and nonpregnant women, and to discuss them, first, in relation to the measurement of estrogens in the urine of women, and second, in the light of recent advances in our knowledge concerning the biosynthesis and metabolism of the estrogens.

Material and Methods

For the purpose of comparing the results of various hydrolytic measures, 12 twenty-four-hour specimens from 3 women who were normally pregnant at 25 weeks to delivery,⁷ and 12 four-day pools from 3 normally menstruating women⁹ were divided into aliquots for various systems of hydrolysis and the extracts assayed both chemically and biologically. In addition to these 24 specimens, the experimental material for chemical versus bioassay to be herein reported was derived from 25 specimens from 3 other pregnancies, from daily collections for 37 days from one normally menstruating woman and for 39 days from another, from 14 collections from normal postmenopausal women, and from 35 other nonpregnancy collections under various pathological or experimental conditions.

All specimens were refrigerated from the start of the collection period and hydrolyzed and extracted within 48 hours of the time the daily collection was completed. Creatinine determinations were made to check on the accuracy of each twenty-four-hour collection. In order to have enough material for accurate bioassay, it was necessary to process extracts of the nonpregnancy urine in two- to five-day pools.

Two systems of hydrolysis were employed on each specimen. One aliquot was hydrolyzed by the two-step glucuronidase plus acid procedure finally adopted for the measurement of the classical estrogens,⁷ viz., refluxed for 10 minutes, adjusted to pH 6.2, incubated for 16 hours at 37° C. with 40 U. of bacterial glucuronidase (Sigma Chemical Company) plus 2.5 mg. of disodium ethylenediamine tetraacetate (Versene) per milliliter of urine, and extracted; the residual urine was then refluxed for 10 minutes with 15 per cent (v + v) concentrated HCl, the extract being combined with that from the glucuronidase procedure. Another aliquot of each was submitted to Zn-HCl hydrolysis,¹² a process long known to alter molecular structure and to be unsuitable for the assay of estrogens as they occur in the urine. Zn-HCl hydrolysis, however, has yielded interesting bioassay results. It was necessary to perform chemical as well as biological assays on the extracts of these aliquots in order to know how much of the estrogenic potency was due to the classical estrogens and how much to unknowns.

Chemical assays (in duplicate) were performed following the method of Brown. His 1955 procedure¹ was used for pregnancy urine. For nonpregnancy urine, in which each color assay is done on the equivalent of a 3 to 4 hour volume, it was found essential, regardless of the method of hydrolysis, to add the purification by saponification step introduced by Bauld¹³ in 1956 and adapted to the Brown method by Brown, Bulbrook, and Greenwood¹⁴ in 1957. Otherwise, underestimates for estriol and overestimates for estrone or estradiol were frequently encountered.

Biological assays were performed on spayed, mature female rats by use of the 50 per cent positive end-point criterion previously described,¹⁰ which requires 24 to 60 rat tests per assay. When the amount of estrogen was too small to permit this degree of accuracy, the negative side of the end point was established and results so expressed, e.g., < 10 R.U./24°.

When chemical and biological assays were compared, the former were converted to rat units on the basis of simultaneously run standardization assays with crystalline estradiol, estrone, and estriol. Bioassays were performed on "total steroid" extracts and at various stages of the fractionation procedures previously described.¹⁰ When unseparated estrogens were bioassayed, the results were compared with the total activity accountable to the chemical assay values for estriol plus estrone plus estradiol. The conversion of micrograms to rat units for this purpose was based on concurrently run standardization assays on

crystalline estradiol, estrone, and estriol combined in the approximate proportions indicated by the chemical assay findings. Separated fractions were compared with standardization assays for the classical estrogen known to be present in each fraction.

The degree of confidence in differences between chemical and bioassay has been assessed by 24 recovery experiments in which estriol, estrone, and estradiol were added to extracts of urine "blanks" (from men and postmenopausal women). In 16 of these experiments,¹⁰ the estrogens were added in the approximate amounts, per 24 hour volume of urine, found in the last trimester of pregnancy. In the other 8 the concentrations were those of normal menstrual cycles.⁹ Chemical assays for the 3 estrogens were performed. Bioassays were done both before and after fractionation. According to the results, there is a 95 per cent degree of confidence in differences between chemical and bioassay of 30 per cent in pregnancy urine, while in nonpregnancy urine differences of 50 per cent or more are significant.

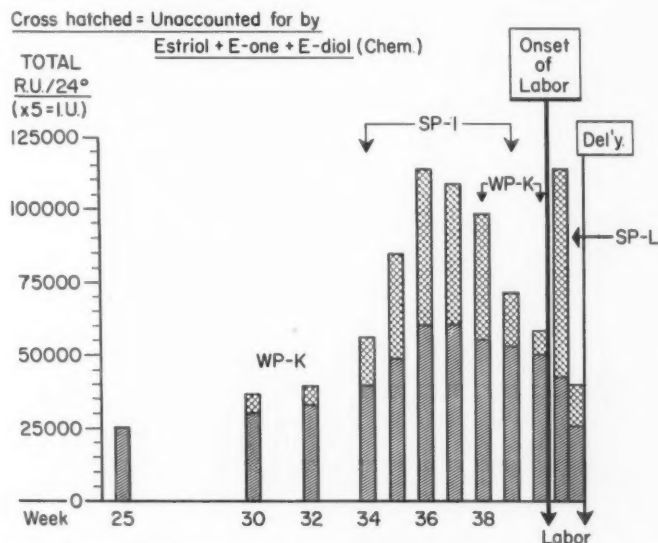


Fig. 1.—Urinary estrogens in a normal pregnancy (glucuronidase plus acid).

Results

Chemical versus bioassay results in pregnancy have been previously published,^{10, 11} it having been these studies which led to the discovery of at least 6 compounds other than estradiol, estrone, and estriol, contributing to the estrogenic potency of the estradiol, estrone, and estriol fractions. In Figs. 1 and 2 the findings on glucuronidase plus acid and Zn-HCl aliquots of urine from a normal pregnancy are summarized, showing how much of the total estrogenic potency was accountable to the classical estrogens and how much to these unknown components.

In the glucuronidase plus acid hydrolyzed aliquots of urine from this patient (Fig. 1), a progressive drop in estriol excretion pertained for 3 weeks prior to the onset of labor and accounted for the relatively slight prepartum decrease in activity accountable to the classical estrogens (chemical assay values converted to rat units). It will be noted that the prepartum drop in total estrogenic activity (bioassay) is considerably more striking. This was due to the

excretion of a strongly phenolic, estriol-like component, designated SP-1, which appeared at 34 weeks, reached peak values at 36 weeks, and fell off rapidly thereafter, none being demonstrable in the specimen collected 3 days before labor. This component is less water-soluble than estriol and may therefore be separated from it. It then has been found to be nonestrogenic per se, but to enhance markedly the biological activity of estriol.¹⁰ That its presence or absence in estriol fractions may be readily detected by bioassay is illustrated in Table VI.

Bioassay of the separated fractions of the glucuronidase plus acid hydrolyzed aliquots revealed, at 30 to 32 weeks and again at term, the presence of a weakly phenolic ketonic component (WP-K), the estrogenic potency of which was of relatively minor significance compared with estriol and SP-1, although many times that of the estrone known to be present in these fractions (see Table V). This estronelike component disappeared when SP-1 was at its highest levels and reappeared prior to the onset of labor.

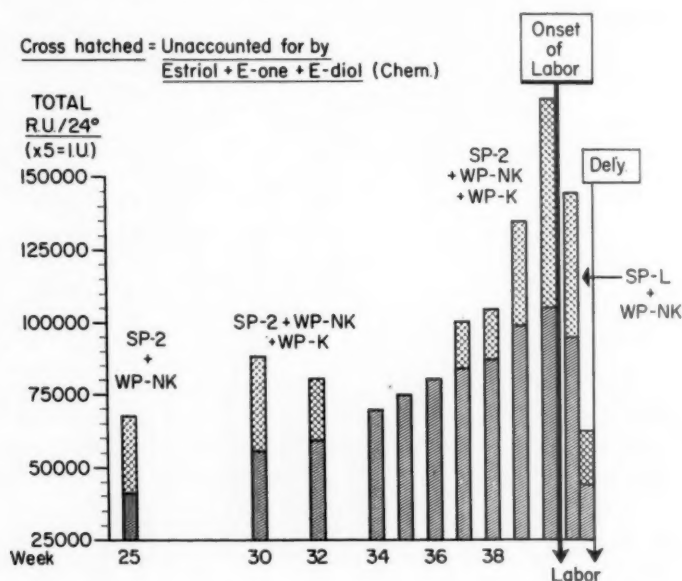


Fig. 2.—Urinary estrogens in a normal pregnancy (Zn-HCl).

During labor, as in our previous studies,^{10, 11} there was a marked drop in the excretion of all 3 of the classical estrogens. This was accompanied by the appearance of an estriol-like component which differed from estriol, from SP-1, and from all unknowns found, in that its estrogenic activity disappeared following the micro-Girard procedure for the separation of ketonic from non-ketonic compounds. It is designated SP-L since, thus far, it has been encountered only in urine during labor.

In the Zn-HCl hydrolyzed aliquots of these same specimens (Fig. 2), the prepartum rise in total estrogenic potency accountable to the classical estrogens was entirely due to an increased recovery of estradiol. From our own studies^{10, 15} and from those of other investigators,¹⁶ the major portion of the estradiol found in Zn-HCl hydrolyzed urine of pregnancy is accountable to the conversion (probably through hydrogenation with zinc and acid) of unknown phenolic components to estradiol. By no means, however, was all of the prepartum rise in these aliquots due to estradiol. An increase in total estrogenic potency *not* accountable to the classical estrogens is very striking during the last 2 weeks of gestation.

In terms of estrogenic potency, the major portion of the unknowns of Zn-HCl hydrolyzed aliquots of normal pregnancy urine is due to the recovery of 2 components which have not been demonstrable in the glucuronidase plus acid hydrolyzed aliquots of the same specimens. One of these, SP-2, is recovered in the strongly phenolic estriol fraction but differs from SP-1 in that it is more water soluble (and therefore more closely allied to estriol) and from SP-L in that it is not labile to treatment with Girard's reagent T, being entirely recovered in the nonketonic fraction. The other, WP-NK, is a weakly phenolic component which is closely allied to estradiol in its solubility characteristics, is nonketonic and stable to Girard's treatment, but differs from estradiol in that it is alkali labile. The other 2 unknowns of Zn-HCl aliquots were also found in the glucuronidase hydrolyzed aliquots of these same specimens; the

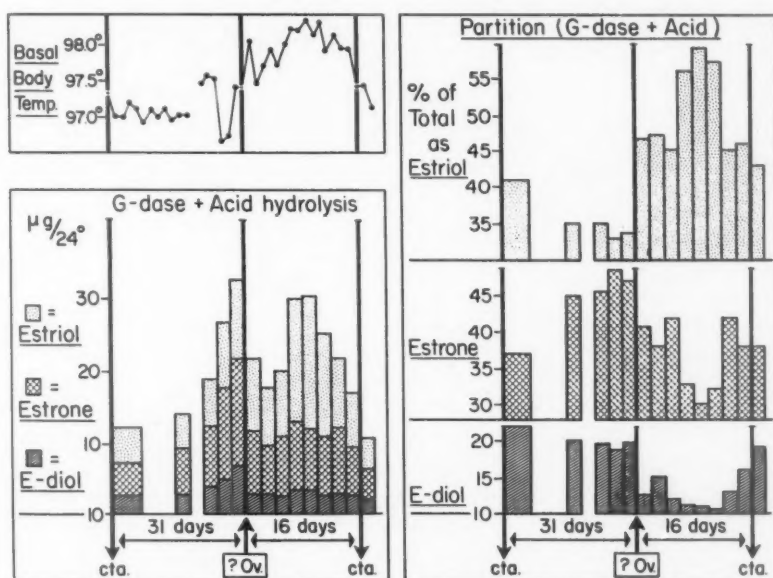


Fig. 3.—Normal menstrual cycle No. 1 (Case 1). Chemical assay.

prepartum estronelike component, WP-K, which was present when SP-1 was low or absent, and small amounts of SP-L, the estriol-like unknown found only during labor.

The most interesting observation in the study of these Zn-HCl unknowns has been the inverse relationship between their curves of excretion and the curves for the excretion of SP-1, the estriol-like unknown which reaches peak values at the 35 to 37 week period of gestation and is found only after glucuronidase or mild acid hydrolysis. It will be noted in Figs. 1 and 2 that the Zn-HCl unknowns disappeared simultaneously with the appearance of SP-1, and that the prepartum drop in SP-1 was accompanied by a progressive increase in Zn-HCl unknowns.

The excretion of the classical estrogens during 2 normal menstrual cycles is shown in Figs. 3 and 4. These curves follow the general pattern of previously reported studies by us^{15, 17} between the years 1938 and 1952 and of the 8 cycles reported by Brown¹⁸ in 1955, although the former were based entirely on bioassay of estradiol, estrone, and estriol fractions and the latter on chemical assay of urines hydrolyzed by refluxing for one hour with acid. There would seem to be little doubt that the time of ovulation is characterized by a peak in estrogen

excretion. This is followed by a drop, a second rise during the luteal phase of the cycle, and a premenstrual fall to levels, during or just after flow, that are the lowest found in normally menstruating women. It is reasonable to assume, as Brown¹⁸ has postulated, that the characteristic drop in the excretion of estrone and estradiol just after the ovulation peak is due to changes in function associated with follicle rupture. This drop, therefore, may be logically assumed to mark the start of the luteal phase. In our studies it has consistently coincided with a rise in basal body temperature to a level above any recorded during the follicular phase.

In Figs. 3 and 4 we have also graphed the partition of the classical estrogens in these 2 normal cycles, that is, the per cent of the total excreted as estriol, estrone, and estradiol, respectively. Coincident with the drop in total estrogens following the ovulation peak, a sudden change in partition is observed with more of the total accountable to estriol and less to estradiol. This pattern is maintained throughout the luteal phase until just before menstruation when it

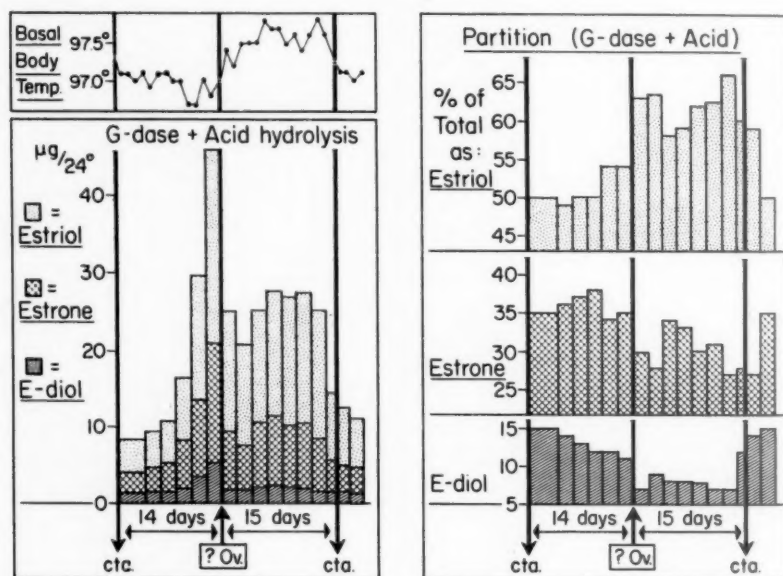


Fig. 4.—Normal menstrual cycle No. 4 (Case 3). Chemical assay.

starts to be reversed, with relatively more estradiol and less estriol. Analyses of our data have shown that this difference between the follicular and luteal phases of normal cycles is statistically significant (Table II). Similar analyses of the cycles reported by Brown in 1955 also revealed a statistically significant relative increase in estriol excretion during the luteal phases of 7 of his 8 subjects, the only exception being in a 40-year-old nullipara whose menstrual flow was abnormally prolonged. This characteristic of the corpus luteum phase of normal cycles is demonstrable, however, only when individual cycles are analyzed. Because there is a wide and overlapping range of normal partition values, composite figures on a number of individuals would reveal no significant differences between phases of the cycle (as was reported by Brown¹⁹). The luteal phase change in estriol excretion in individual cases, however, is quite clear, and appears to be absolute as well as relative. Because of the wide range throughout the cycle of absolute values (i.e., micrograms per 24 hours), the 2 phases cannot be statistically compared in this respect, but the average daily excretion of estriol, as well as of total estrogen, was higher after ovulation than

before in all 3 of our subjects (Table II), and in 7 of the 8 cycles reported in 1955 by Brown¹⁸ (the same 7, incidentally, in which partition values showed a relative increase in estriol).

Chemical versus bioassay results in these 2 normal cycles are presented in Figs. 5 and 6, in which the total estrogenic potencies of the glucuronidase plus

Cross hatched = Unaccounted for by Estriol + E-one + E-diol (Chem.)

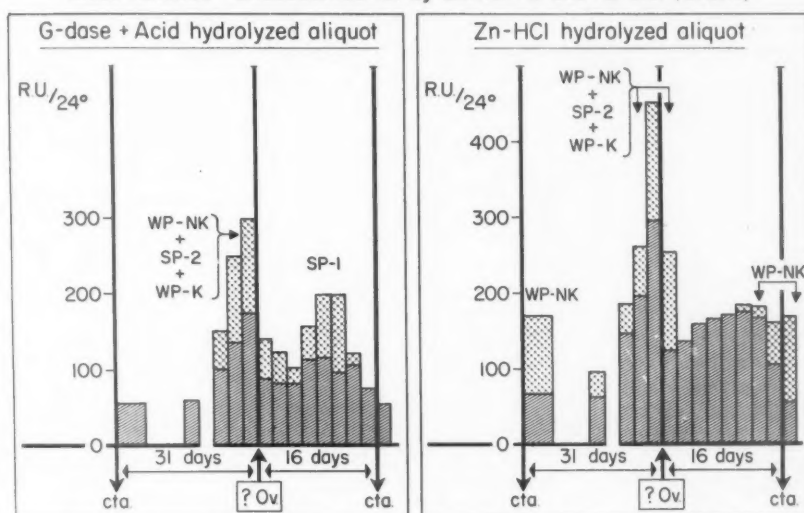


Fig. 5.—Normal menstrual cycle No. 4 (Case 1). Biological assay.

Cross hatched = Unaccounted for by Estriol + E-one + E-diol (Chem.)

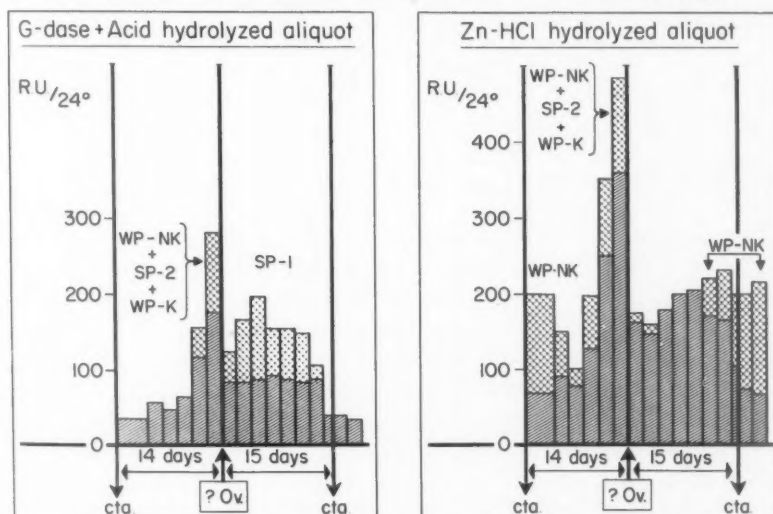


Fig. 6.—Normal menstrual cycle No. 4 (Case 3). Biological assay.

acid and Zn-HCl hydrolyzed aliquots are compared with their total activities accountable to the classical estrogens. As in normal pregnancy, more estrogen was often found in both aliquots than could be accounted for by the estriol, estrone, and estradiol known to be present, and the curves of excretion followed

definite patterns related to the phase of the cycle. In order to have enough estrogen for accurate bioassay after fractionation (a necessary step in the characterization of unknowns) all that was left of our normal cycle extracts (after the bioassays for total activity) were combined in 3 pools representing the menstrual phase, the ovulation time peak, and the luteal phase maximum, respectively. In order to illustrate how the presence or absence of the various unknown components may be detected by chemical versus bioassay, representative findings in separated fractions are presented in Tables III-VI.

TABLE II. URINARY ESTRIOL, ESTRONE, AND ESTRADIOL IN NORMAL OVARIAN CYCLES; PARTITION IN THE FOLLICULAR AND LUTEAL PHASES

CASE	PHASE OF CYCLE	NO. OF OBSERVATIONS	AVERAGE TOTAL $\mu\text{G}/24^\circ$ ($E_1 + E_2 + E_3$)	PER CENT OF TOTAL					
				ESTRIOL		ESTRONE		ESTRADIOL	
				AVERAGE \pm STANDARD DEVIATION	P*	AVERAGE \pm STANDARD DEVIATION	P*	AVERAGE \pm STANDARD DEVIATION	P*
1	Follicular†	6	19.1	37 \pm 4		43 \pm 5		20 \pm 1.3	
	Luteal†	11	22.3	50 \pm 5	(0.01)	37 \pm 4	(0.05)	13 \pm 1.8	(0.01)
2	Follicular	2	25.8	35 (34 & 36)		37 (34 & 40)		28 (32 & 24)	
	Luteal	1	30.2	48		34		18	
3	Follicular	10	16.0	51 \pm 3		36 \pm 4		13 \pm 1.6	
	Luteal	9	24.6	61 \pm 3	(0.01)	31 \pm 3	(0.01)	8 \pm 1.7	(0.01)
Average per cent change (luteal vs. follicular)			+29%	+31%		-12%		-36%	

*Significance of difference between follicular and luteal phase.

†Follicular = day 1 through ovulation peak in total micrograms per 24 hours. Luteal = rest of cycle to start of next menstrual flow.

TABLE III. CHEMICAL VERSUS BIOASSAY OF ESTRADIOL FRACTIONS CONTAINING WP-NK COMPARED WITH SIMILAR FRACTIONS IN WHICH IT WAS ABSENT

CLINICAL NOTES ON URINE DONORS	METHOD OF HYDROLYSIS†	CHEMICAL R.U. $\text{C}/24^\circ$ ESTRADIOL	BIOASSAY (R.V./24°)	
			TOTAL E-DIOL + "X"	DIFFERENCE* "X"
<i>Pregnant.</i> —				
Normal during labor	(Z)	35,100	51,000	15,900
	(G)	2,900	3,250	0
Same case at 37 weeks	(Z)	37,100	32,000	0
	(G)	4,500	4,250	0
<i>Normal Cycle Pools.</i> —				
Menstrual phase	(Z)	93	160	67
	(G)	40	35	0
Ovulation peak	(Z)	250	380	130
	(G)	98	200	102
Luteal phase maximum	(Z)	108	120	0
	(G)	60	70	0
<i>Postmenopausal.</i> —				
Cancer of breast \bar{c}	(Z)	0	2,000	2,000
ovarian C.S.H.‡	(G)	0	6,450	6,450
Same case after oophorectomy	(Z)	208	200	0
	(G)	70	80	0
<i>Adrenal Adenoma.</i> —				
Before removal of tumor	(Z)	160	320	160
	(G)	80	237	157
Same case postoperative	(Z)	78	160	82
	(G)	59	63	0

*Any difference below the level of a 95 per cent degree of confidence in differences between chemical and bioassay results is listed as zero in this and the following tables (see Methods).

†Z = Zn-HCl hydrolyzed aliquots; G = glucuronidase hydrolyzed aliquots.

‡Four- to five-day pools of urine were analyzed from these donors.

§C.S.H. = cortical stromal hyperplasia.

TABLE IV. CHEMICAL VERSUS BIOASSAY OF ESTRIOL FRACTIONS CONTAINING SP-2*
COMPARED WITH SIMILAR FRACTIONS IN WHICH IT WAS ABSENT

CLINICAL NOTES ON URINE DONORS	METHOD OF HYDROLYSIS†	CHEMICAL (R.U. \sim /24°) ESTRIOL	BIOASSAY (R.U./24°)	
			TOTAL ESTRIOL + "X"	DIFFERENCE† "X"
<i>Pregnant.</i> —				
Normal at term	(Z)	56,400	97,000	40,600
	(G)	57,100	67,000	0
Same case at 37 weeks	(Z)	41,280	39,500	0
	(G)	39,750	41,000	0
<i>Normal Cycle Pools.</i> —				
Menstrual phase	(Z)	10	10	0
	(G)	8	10	0
Ovulation peak	(Z)	30	55	20
	(G)	34	60	26
Luteal phase maximum	(Z)	35	40	0
	(G)	40	45	0
<i>Postmenopausal.</i> —				
Cancer of breast \bar{c} ovarian C.S.H.	(Z)	6	98	92
	(G)	8	160	152
Same case after oophorectomy	(Z)	9	< 10	0
	(G)	15	20	0
Surgical castrate \bar{c} cancer of breast	(Z)	10	75	65
	(G)	8	< 10	0
Same case after adrenalectomy	(Z)	3	< 5	0
	(G)	3	< 5	0
Surgical castrate on 5 mg. estrone daily	(Z)	1,300	1,950	650
	(G)	1,230	1,250	0
Same case on estrone + pro- gesterone	(Z)	1,130	1,250	0
	(G)	1,420	1,200	0
<i>Adrenal Adenoma.</i> —				
Before removal of tumor	(Z)	21	38	17
	(G)	24	90	72
Same case postoperative	(Z)	22	20	0
	(G)	21	23	0

*Estriol fractions separated by water out of benzene-petroleum ether 1:1.

†See footnotes to Table III.

Analyses after fractionation revealed that the menstrual phase was accompanied by the excretion of only one unknown, an estradiol-like, WP-NK component. As in pregnancy urine at term and during labor (Fig. 2 and Table III), this was found after Zn-HCl hydrolysis but not after glucuronidase plus acid. There were no unknowns in glucuronidase plus acid hydrolyzed aliquots of menstrual urine (Figs. 5 and 6, Tables III-VI).

At the time of ovulation the differences between chemical and bioassay results were found to be due to the excretion of 3 different components. The major constituent in terms of estrogenic potency was an estradiol-like unknown. This WP-NK component, unlike that of menstrual and pregnancy urines, was recovered in both aliquots—the glucuronidase plus acid as well as the Zn-HCl hydrolyzed (Table III). The second component at the time of ovulation was a substance similar to the SP-2 of pregnancy urine in that it was water-soluble, like estriol, and stable to Girard's separation, being entirely recovered in the nonketonic fraction. At ovulation time, however, the component with these characteristics was found in the glucuronidase hydrolyzed aliquot (Table IV) as well as in the Zn-HCl pool, whereas an unknown with the characteristics of SP-2 has not been demonstrable by bioassay in glucuronidase hydrolyzed urines of normal pregnancy. The third unknown from urines collected at ovulation

time was an estronelike, WP-K substance which was found both after Zn-HCl and glucuronidase hydrolysis and was thus entirely similar to the WP-K component of pregnancy urine (Figs. 1 and 2 and Table V).

TABLE V. CHEMICAL VERSUS BIOASSAY OF ESTRONE FRACTIONS CONTAINING WP-K COMPARED WITH SIMILAR FRACTIONS IN WHICH IT WAS ABSENT

CLINICAL NOTES ON URINE DONORS	METHOD OF HYDROLYSIS*	CHEMICAL (R.U. ∞ /24°) ESTRONE	BIOASSAY (R.U./24°)	
			TOTAL ESTRONE + "X"	DIFFERENCE* "X"
<i>Pregnant.</i> —				
Normal at term	(Z)	160	6,100	5,940
	(G)	1,430	6,800	5,370
Same case at 37 weeks	(Z)	88	< 100	0
	(G)	1,140	1,060	0
<i>Normal Cycle Pools.</i> —				
Menstrual phase	(Z)	2	< 5	0
	(G)	7	< 10	0
Ovulation peak	(Z)	4	40	36
	(G)	20	40	20
Luteal phase maximum	(Z)	2	< 5	0
	(G)	16	19	0
<i>Postmenopausal.</i> —				
Cancer of breast & ovarian	(Z)	3	100	97
C.S.H.	(G)	5	35	30
Same case after oophorec-	(Z)	3	< 5	0
tom y	(G)	8	< 10	0
<i>Adrenal Adenoma.</i> —				
Before removal of tumor	(Z)	2	20	18
	(G)	13	70	57
Same case postoperative	(Z)	2	< 5	0
	(G)	11	10	0

*See footnotes to Table III.

TABLE VI. CHEMICAL VERSUS BIOASSAY OF ESTRIOL FRACTIONS CONTAINING SP-1* COMPARED WITH SIMILAR FRACTIONS IN WHICH IT WAS ABSENT

CLINICAL NOTES ON URINE DONORS	CHEMICAL (R.U.-/24°) ESTRIOL	BIOASSAY (R.U./24°)	
		TOTAL ESTRIOL + "X"	DIFFERENCE† "X"
<i>Pregnant.</i> —			
Normal at 36 weeks	49,600	98,600	49,000
Same case at term	39,500	38,000	0
Normal at 31 weeks	10,800	20,000	9,700
Same case at 33 weeks, just before premature labor	7,500	9,100	0
<i>Normal Cycle Pools.</i> —			
Menstrual phase	8	10	0
Ovulation peak	"X" estriol = SP-2 (see Table IV)		
Luteal phase maximum	40	85	45
<i>Postmenopausal.</i> —			
Surgical castrate on 5 mg. estrone daily	1,230	1,060	0
Same case on estrone + progesterone	1,420	3,500	2,080
Surgical castrate on 150 mg. of estriol daily for 4 days	65,000	106,000	41,000
Control urine of same patient plus 10 mg. estriol per 24 hour volume	17,900	18,500	0

*Estriol fractions separated by N/10th NaOH out of ether. Found only in glucuronidase and 10 minute acid hydrolyzed aliquots.

†See footnotes to Table III.

During the midluteal phase of the cycles studied there was no evidence in the Zn-HCl aliquots for estrogenic substances other than estriol, estrone, and estradiol. This was true in the individual collections assayed before separation (Figs. 5 and 6) and also in analyses after fractionation of the luteal phase maximum pool (Tables III-V). In the glucuronidase plus acid hydrolyzed aliquots, on the other hand, more estrogen was revealed by bioassay than could be accounted for by the classical estrogens. All of this difference was traced to SP-1 (Table VI), the estriol-like unknown recovered when estriol is separated from the neutral and weakly phenolic steroids by extraction out of ether with 0.1 normal NaOH, but is not recovered by extraction of the estriol fraction out of benzene-petroleum ether with water, the procedure that recovers SP-2. The fact that SP-1 is excreted only during the luteal phase of normal cycles, and only at the 34 to 38 week period of pregnancy (Fig. 1), when a number of lines of evidence point to a peak in placental function, makes it of particular interest. It is also of interest that the other unknowns, in normal cycles as in pregnancy, are not recovered when SP-1 is being excreted in considerable amounts.

Observations concerning chemical versus bioassay results for urinary estrogens in anovulatory situations before and after the menopause will be reported in more detail elsewhere. A few representative findings are included in Tables III-VI, and will be commented upon briefly.

On the basis of bioassay, abnormally high estrogen output in *postmenopausal women with cancer of the breast* has been reported by several investigators.²⁰⁻²² Studies in this laboratory have shown rising titers with advancing disease and a fall to normal levels shortly after oophorectomy.^{22, 23} In all of 8 postmenopausal women with breast cancer whose ovaries were later found to show pronounced cortical stromal hyperplasia, urinary estrogens were markedly elevated.²³ In the studies of 4 of these women with very high titers, chemical as well as bioassays were performed. The excretion of the classical estrogens in 3 of the 4 was found to be within the limits of normal for postmenopausal women. Bioassay of the separated fractions revealed that most of the estrogenic activity was accountable to an estradiol-like unknown, which, like the WP-NK of urines excreted by normal women at the time of ovulation, was recovered after glucuronidase as well as Zn-HCl hydrolysis. In one patient (Table III) the estrogenic potency of the estradiol fraction of the glucuronidase hydrolyzed aliquot was as high as that found in the second trimester of normal pregnancy, whereas chemical assay revealed that no estradiol was present, and only 4 and 2 μ g, respectively, of estriol and estrone per 24 hour volume. Two other unknowns contributed, to a lesser extent, to the estrogenic potencies of the urines of these 4 women prior to oophorectomy, an estriol-like component with the characteristics of SP-2, and an estronelike substance with the characteristics of WP-K (Tables IV and V). Both were recovered in glucuronidase as well as in Zn-HCl hydrolyzed aliquots, thereby resembling again the unknowns excreted by normal women at the time of ovulation. Soon after oophorectomy these 3 unknowns disappeared and the bioassay titers returned to normal levels (Tables III-V).

In *oophorectomized women with advancing metastatic cancer* of the breast, however, high and rising titers for estrogenic potency have also been observed, especially in Zn-HCl hydrolyzed aliquots.²² Chemical assays on glucuronidase hydrolyzed aliquots of such urines have revealed an unusual partition of the classical estrogens with 50 per cent or more of the total, which is not necessarily high, excreted as estradiol. The estradiol content of Zn-HCl hydrolyzed aliquots has been considerably higher, but not enough to account for the bioassay findings. Bioassays on separated fractions have revealed that the major portion of the estrogenic potency of Zn-HCl aliquots in this situation is accountable to an estriol-like component with the characteristics of SP-2, but, unlike the SP-2 of women with ovaries, found only after Zn-HCl hydrolysis. In one such patient

urinary estrogens were again studied by both chemical and bioassay after adrenalectomy (Table IV). No SP-2 was recovered in either aliquot, and the total estradiol, estrone, and estriol (chemical assay) was also very low with a total of only 1.7 μg per 24 hour volume (in comparison with 6.6 μg per 24 hours before operation).

In a patient with amenorrhea due to an *adrenal adenoma* (whose urinary 17-ketosteroids were over 200 mg. per 24 hours), urinary estradiol, estrone, and estriol were within normal limits and remained unchanged after removal of the tumor. In 3 collections (4 to 5 day pools) over a period of one month before operation, the total output of the classical estrogens was 18.6 to 21.0 μg per 24 hours in the glucuronidase plus acid hydrolyzed aliquots. This patient had at that time been amenorrheic for 5 months. In a 5 day postoperative collection just before and at the start of the first menstrual period (days 16-20 after removal of the tumor) 20.6 μg of estradiol plus estrone plus estriol were excreted per 24 hour volume with essentially no change in the relative proportions of each. There was a considerable difference between the pre- and postoperative specimens, however, in their estrogenic potencies, both as to total rat units per 24 hours, and the characteristics of the unknowns. The preoperative glucuronidase plus acid hydrolyzed aliquots contained 330 to 390 R.U. per 24 hours. On days 16 through 20, after removal of the tumor, 96 R.U. per 24 hours was excreted. Bioassay of the separated fractions revealed the presence of 3 unknowns before operation: an estradiol-like component with the characteristics of WP-NK, an estriol-like SP-2 factor, and an estronelike WP-K compound (Tables III-V). All 3 were recovered in Zn-HCl as well as glucuronidase hydrolyzed aliquots. As may be seen in the tables, none of them was found in the glucuronidase aliquots of the postoperative collection made just before and at the start of the patient's first menstrual period. The only unknown at that time was an estradiol-like WP-NK component which, as in the urine of normal women at the time of menstruation, was recovered only after Zn-HCl hydrolysis (Table III).

SP-1, the estriol enhancer which characterizes the luteal phase of the normal cycle and the 34 to 38 week period of gestation, has not as yet been encountered as a natural product in the urine of anovulatory women either before or after the menopause. It has been found, however, in the urines of *postmenopausal women under experimental conditions*. A normal woman, whose ovaries had been removed surgically, was given 5 mg. of estrone daily by mouth for 20 days (in the form of estrone sulfate) plus 400 mg. of buccal progesterone daily on days 7 through 12 of the experimental period. A changed partition of the classical estrogens was observed during and just after the 6 days of progesterone administration, with a relative increase in estriol output. There was no significant increase in the total output of estriol plus estrone plus estradiol on progesterone, but the total estrogenic potency of glucuronidase plus acid hydrolyzed aliquots was doubled while progesterone was being administered. All of this increase was found, upon bioassay after fractionation, to be due to the excretion of SP-1 (Table VI). This estriol enhancer also appeared in the urine of 3 postmenopausal women given 150 mg. of estriol daily by mouth for 4 days (Table VI).⁷ The excretion of SP-1, therefore, appears to be associated with various situations in which progesterone is present and/or a relatively high level of estriol is being excreted.

In the woman who had undergone oophorectomy and who was given estrone with and without progesterone, the Zn-HCl hydrolyzed aliquots contained no unknowns during the period of progesterone administration. When she was on estrone only, however, an estriol-like component with the characteristics of SP-2 was recovered in these aliquots (Table IV). Since none was found in the glucuronidase hydrolyzed aliquots of the same specimens, this SP-2 component

resembled the SP-2 of pregnancy urine (Fig. 2) rather than the compound with similar solubility characteristics found at ovulation time in both glucuronidase and Zn-HCl hydrolyzed urine (Table IV).

Comment

The Measurement of Estrogens in the Urine of Women as a Clinical Procedure.—The findings herein reported have added to, without identifying the number of physiologically important urinary estrogens and made only more difficult the problem of a practical means of measuring estrogens in the urine of women as a guide to diagnosis and treatment. For the time being, bioassay would seem the only solution, but it is not practical since reliable results can be obtained only by a time-consuming process involving the use of 24 to 60 animal tests per assay.

Chemical assay, by the Brown procedure, is informative as regards the classical estrogens and this information alone is of considerable value in the study of *pregnant* or *premenopausal* patients, but only if repeated assays are performed on each case studied. With use of Brown's method (with our modification as to hydrolysis), an abnormal drop in estriol prior to spontaneous premature labor has been observed.¹¹ This drop, as in the case of spontaneous labor at term herein reported (Fig. 1), was considerably less striking than that in the estriol-like unknown, SP-1, which can be measured only by bioassay. It is now apparent that we were not measuring just estriol in our early studies in which the rate of excretion of "estriol" appeared to provide a more sensitive index of placental function than could be acquired by any other hormonal measurements (e.g., chorionic gonadotropin or urinary pregnanediol).²⁴ The marked deviations from the normal that were found to precede late pregnancy complications must have been contributed to largely by the rate of excretion of SP-1, the estriol enhancer which characterizes the 34 to 38 week period of normal pregnancy and the luteal phase of the normal cycle. However, since the curve of excretion of SP-1 follows (and exaggerates) the curve of estriol excretion, any situation involving premature placental senescence presumably would be accompanied by a drop in estriol output. Zondek,²⁵ who bases his conclusions on a chemical assay procedure, recently has suggested again that the rate of estriol excretion in late pregnancy closely reflects placental function. Taylor and associates,²⁶ also on the basis of a chemical assay procedure, have confirmed our early reports that dropping estrogens, rather than normally rising titers, are indicative of premature placental failure. The range of normal values for urinary estrogens in pregnancy is so great, however, that prognostic value, in an individual case, can be attached only to assays that are repeated sufficiently often to reflect the curve of excretion.

The same situation holds in the study of patients with *menstrual disorders*. Because of the wide range of normal values, an individual situation could be assessed only by frequent collections over a period of weeks or even months. This is apparent from the studies of normal cycles herein reported, and has been Brown's experience in studies of normal cycles^{18, 27} and menstrual disorders.²⁸ Brown's method of chemical assay is not a simple process and it requires not only time but extremely accurate quantitative techniques. Since the rate of estriol excretion seems to be of especial significance both in the ovarian cycle and in pregnancy, it is possible that a simplified system for the chemical assay of just estriol might give informative results. Such a system has been proposed by Eberlein and associates,²⁹ whose findings during a normal menstrual cycle look promising.

In our experience the use of chemical assays for the study of *postmenopausal women* has failed to give results at all indicative of the metabolic changes so

strikingly shown by bioassay. In postmenopausal women with breast cancer and high bioassay titers, for example, oophorectomy has been consistently followed by a pronounced drop in the estrogenic potency of the urine. This has not been accompanied by any consistent changes in the output of the classical estrogens, an observation in keeping with the much more extensive experience, with the Brown method, in the study of such cases by Bulbrook and co-workers.^{30, 31} These investigators concluded, in fact, that neither oophorectomy nor adrenalectomy has any consistent effect on the urinary excretion of estrogens by postmenopausal women. A rise in titer after these operative procedures has been as frequently observed as a fall and is unrelated to the preoperative levels, most of which are at the lower limits for the precision and sensitivity of chemical assay. Paulsen and associates,^{32, 33} who use bioassay only, on the other hand, have obtained evidence for a close correlation between estrogen output and the presence or absence of the ovaries in postmenopausal women. In postmenopausal

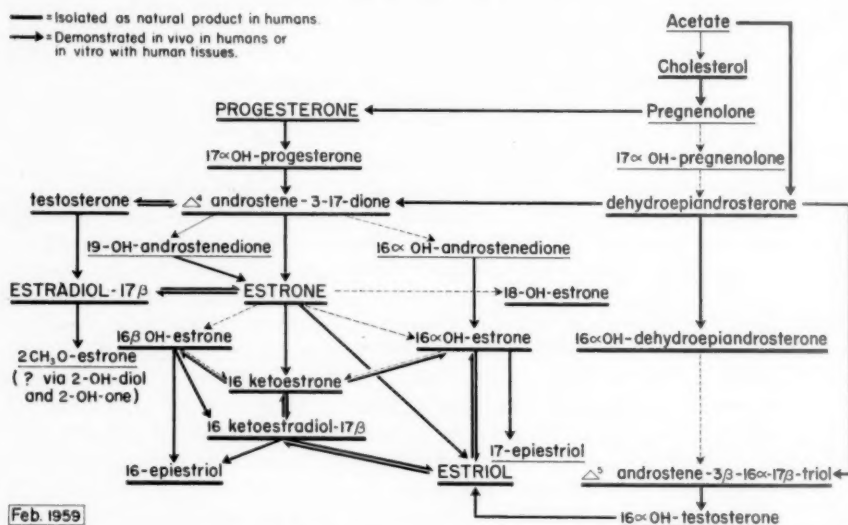


Fig. 7.—Pathways of estrogen biosynthesis and metabolism. Solid lines represent reactions in which the end products have been chemically characterized. Dotted lines represent pathways that are theoretically possible. (The references upon which this figure is based will be supplied by the authors upon request.)

women it would appear that estrogens other than estradiol, estrone, and estriol are the major urinary constituents when high titers are encountered. Until these unknowns have been chemically identified their presence can be detected only by bioassay.

Unknown Urinary Estrogens in the Light of Recent Advances in Our Knowledge Concerning the Biosynthesis and Metabolism of the Estrogens.—Seven phenolic compounds other than estradiol, estrone, and estriol have now been identified as natural products in human urine, and an eighth has been shown to be a metabolic product of one of these by in vitro studies with human liver (Fig. 7). All of these compounds except one, 16-ketoestrone, are Kober chromogens. Since they do not interfere with the specificity of Brown's assay for estradiol, estrone, and estriol,²⁷ they might be responsible for our findings. We have evidence, however, that the estriol-like unknowns, as they are recovered from pregnancy urine where they are the major source of unaccounted-for estrogenic potency, are not Kober chromogenic.⁷ Three of the newer phenolic compounds, 16-ketoestrone, 16-ketoestradiol, 17β and 2-methoxyestrone, are either non-estrogenic or of very low estrogenic potency, but might be converted into

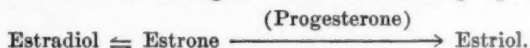
estrogenic forms by Zn-HCl hydrolysis. We have in fact found that Zn-HCl hydrolysis of 16-ketoestrone, of 16-ketoestradiol, and of the triacetate of 2-hydroxyestradiol added to urine "blanks" results in a marked increase in their respective estrogenic potencies, most of which cannot be attributed to the formation of any of the classical estrogens (unpublished findings). No 16 α hydroxyestrone has been available to us, but it might similarly be changed by treatment with zinc and acid into a more estrogenic form. A recent contribution by Breuer and associates,³⁴ in which 16-ketoestrone was incubated with human liver and ovaries, confirms the earlier in vivo studies of Stimmel, Grollman, and Huffman,³⁵ and indicates that 16-ketoestrone and 16-ketoestradiol may be key compounds among the intermediaries involved in the C-16 hydroxylation of estrone (Fig. 7). Our findings in Zn-HCl hydrolyzed aliquots have indicated that the estrogens measured therein provide a gauge of intermediary estrogen metabolism, as opposed to rapid conversion to estriol.

In normal women high levels of Zn-HCl unknowns pertain before the onset of spontaneous labor and at the menstrual and ovulatory phases of the ovarian cycle. When the ovaries are relatively quiescent, as in late pregnancy and at the start of menstruation, such unknowns are recovered only after Zn-HCl hydrolysis. In the normal cycle, unknowns with these characteristics are particularly prominent at the time of ovulation, when they appear in glucuronidase as well as Zn-HCl hydrolyzed aliquots. In recent studies by Brown and co-workers,²⁷ in which urinary estrogens and gonadotropins were measured daily during 9 menstrual cycles, it was noted that the midcycle rise in gonadotropins never preceded the estrogen peak. In 4 cases the gonadotropin rise came after that in estrogens, in 3 they coincided, and in the other 2 women, who had poor ovulation peaks in estrogen excretion, there was no midcycle rise in gonadotropins. This, of course, suggests that the estrogen peak might be causally related to the midcycle increase in gonadotropin excretion. The fact that the marked rise in output of the classical estrogens at this time is accompanied by an equally striking increase in the excretion of biologically active material with the characteristics of the Zn-HCl unknowns is of particular interest in connection with our early observations and resultant concepts concerning the retroactive effects of intermediary metabolites of the estrogens upon the pituitary.^{15, 24}

That pregnenolone, progesterone, 17 α -hydroxyprogesterone, and androstenedione may be important intermediaries in the biosynthesis of the estrogens from cholesterol is now well documented (Fig. 7). Recent evidence, derived from studies by Ryan^{36, 37} on the metabolism of C-16 oxygenated C₁₉ steroids by human placental enzymes, indicates that estrone and estradiol are not necessarily precursors in the formation of estriol. He has shown the conversion of 16 α -hydroxyandrostenedione to estriol by way of 16 α -hydroxyestrone, and of Δ^5 -androstene-3 β -16 α -17 β -triol to estriol by way of 16 α -hydroxytestosterone (Fig. 7). The Δ^5 -triol compound is a known constituent of normal urine.³⁸ It is commonly referred to as Hirschmann's triol since it was first found by him, in very large amounts, in the urine of a patient with an adrenal tumor.³⁹ The 16 α -hydroxy derivative of androstenedione, which might be a metabolite of progesterone, has not been identified in human urine, but is closely related in its structure to compounds known to be normally excreted.⁴⁰ Neither of these C₁₉ steroids is estrogenic (unpublished observation), but we have just recently discovered that a 1:1 combination of 16 α -hydroxyandrostenedione with estriol gives an 80 per cent enhancement of estriol's estrogenic activity. Moreover, we have found that this C₁₉ steroid is partially taken into the strongly phenolic estriol fraction when an ether extract of the phenolic plus neutral steroids is extracted with 0.1 NaOH (the procedure which recovers SP-1, the estriol enhancer) and, like SP-1, is not extracted out of benzene:petroleum ether with water (thereby differing from estriol and SP-2). The fact that the excretion of SP-1 is so closely correlated

with situations associated with high levels of progesterone and relatively high estriol output is of particular interest. It is tenable to suggest that SP-1, as well as estriol, could sometimes be derived from progesterone without the intermediate formation of estrone and estradiol.

The schematic presentation in Fig. 7 of our present state of knowledge concerning the biosynthesis and metabolism of the estrogens depicts 3 possible pathways for the formation of estriol and may explain a number of paradoxes in observations concerning estriol excretion. At the 1938 meeting of this Society a very simple presentation of estrogen metabolism was proposed⁴¹:



This was based on experimental observations by Pincus and Zahl^{42, 43} in rabbits and by ourselves^{41, 44} in rabbits and women. It was later confirmed in women by Pincus and Graubard.²⁰ All of these observations depended upon bioassay of urinary estrogens, except that Pincus also used the David test, a highly specific color reaction for estriol. That progesterone caused increased excretion of total estrogens in a woman with ovarian agenesis, with an absolute and relative increase in estriol output, was also reported by Engel⁴⁵ in 1951. This was based on countercurrent distribution. In 1953, however, Pearlman and co-workers⁴⁶ reported studies with deuterium-labeled estrone in which they found no difference between pregnant and nonpregnant women in the conversion of estrone to estriol. These experiments appeared to provide conclusive evidence that progesterone was not concerned in estriol production, a conclusion which must now again be questioned, both on the basis of the data presented herein and the recent findings of other investigators. The very large amounts of estriol excreted in late pregnancy and the fact that it accounts for about 90 per cent of the total excretion of the classical estrogens from the second trimester to term have long aroused speculation as to its source. The demonstration by Ryan of enzymes in the placenta capable of synthesizing estriol directly from C₁₉ steroids is indeed an important contribution. It remains to be determined whether *in vitro* findings are applicable to *in vivo* situations. In any event, however, our rapidly advancing knowledge concerning the enzyme systems which may be involved in the biosynthesis and metabolism of the estrogens has opened up new vistas, the exploration of which may in time explain the findings herein reported.

Summary

A systematic study of various hydrolytic procedures has revealed that incubation of urine with bacterial glucuronidase followed by extraction and mild acid hydrolysis of the residual urine consistently gives higher yields of estradiol, estrone, and estriol from the urine of women than are recovered by any single-step measure. In Brown's procedure for the measurement of these 3 estrogens (the chemical assay employed in this investigation) the urine is hydrolyzed by boiling for one hour with 15 per cent HCl. Because of the comparatively low recoveries and lack of uniformity of results with this measure, his method was modified by substituting glucuronidase followed by 10 minute acid hydrolysis. This change did not detract from the accuracy, sensitivity, and specificity of Brown's chemical assay of the 3 classical estrogens.

Primarily as a check on the specificity of the chemical assay, each extract was also bioassayed both before and after fractionation. An offshoot of this part of the work, and the subject of the present communication, has been the discovery that unknown components of the phenolic fractions contribute as significantly to the estrogenic potencies of the urines of women as do estriol, estrone,

and estradiol. The types of unknowns found and their rates of excretion follow definite patterns in relation to known placental changes in pregnancy and to the stage of ovarian activity in the nonpregnant.

One of these unknowns, an estriol-like component designated SP-1, reaches its highest levels 2 to 4 weeks before term in normal pregnancy, falls off rapidly thereafter, and disappears prior to the onset of labor. In normal cycles it appears only during the luteal phase, reaching peak values midway between ovulation and the start of the next period. It has never been found as a natural product in the urine of anovulatory or postmenopausal women. In a woman whose ovaries were removed and who was given estrone with and without progesterone, it appeared only while progesterone was being administered. It was also found in the urine of 3 postmenopausal women given 150 mg. of estriol daily for 4 days.

The other unknowns, of which there are at least 5 (according to differences in their solubility characteristics and lability to various steps in the fractionation procedures), have not been encountered in the urine during those physiological states associated with high progesterone secretion and the accompanying relatively high levels of urinary estriol. One of them, an estriol-like component that is destroyed by Girard's separation, has been found only during labor; and another, an acidic component, only just before the onset of labor. The other 3 are common constituents of both pregnancy and nonpregnancy urines. These 3 reach peak values just before the onset of labor and just before ovulation in normal cycles. They are often the only estrogens excreted in appreciable amounts by postmenopausal women with mammary cancer, and account for the very high titers associated with ovarian cortical stromal hyperplasia. In a woman with an adrenal adenoma whose urinary estradiol, estrone, and estriol were normal and unchanged by removal of the tumor, these 3 components were responsible for 75 per cent of the estrogenic activity of the urine before operation. In abnormal situations, therefore, estrogens other than estradiol, estrone, and estriol are often the only ones that differ from the normal.

The significance of these findings is discussed, first, in relation to the measurement of estrogens in the urine of women, and second, in the light of recent advances in our knowledge concerning possible pathways in the biosynthesis and metabolism of the estrogens.

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Discussion

DR. ERNEST W. PAGE, San Francisco, Calif.—To me, the most interesting clinical facts brought out in this paper are these: (1) there are one or two nonestrogenic steroids in human urine which are capable of enhancing the biologic effect of the classical estrogens; at least one of these is a product of progesterone and may be 16- α -hydroxyandrostenedione; (2) this compound, plus the weakly phenolic ketonic material which appears to replace it in the last 2 weeks of pregnancy, may be related in some way to the onset of labor; (3) estriol may be formed from pregnenolone without passing through estrone or estradiol; (4) postmenopausal women with breast cancer and ovarian cortical stromal hyperplasia have a high urinary excretion of unknown estrogens; (5) the essayists present us with valuable data pertaining to the excretion of estrogens in the normal menstrual cycle and during normal pregnancy.

With respect to the last point, I would like to present some preliminary data on the elaboration of estrogens by the isolated placenta. We have all assumed correctly that the placenta is the major source of estrogens in pregnancy, but direct proof has been lacking. Steroid hormones are not produced in placental tissue cultures, and the extraction of hormone from an organ does not always reveal the original source of the hormone. We have been doubly perfusing the fetal and maternal sides of the placenta for several years, but have only recently directed our attention to its estrogen production. Thus far, we have merely

determined biologic activity, expressed as equivalents of estrone, but my co-worker, Dr. Glendening, is proceeding with the Brown method for classical estrogens and in the near future we should have more precise data.

The perfusate consists of salt solutions with human albumen and tissue culture nutrients added. The average estrogen production of 8 placentas is shown in Table I.

TABLE I

NO.	PERFUSATE	ESTRONE EQUIVALENTS (μ G/KG./HR.)	CHORIONIC GONADO- TROPIN (MOUSE UNITS/L.)
8	Albumen plus nutrients	270	500- 2,000
2	Albumen alone	50	5,000-10,000
2	Nutrients alone	0	500- 5,000

It is of interest to note that when the special nutrients are omitted, estrogen production is markedly reduced and that when albumen is omitted, no hormone release can be detected. Dr. Olive Smith will be pleased to note that the reduced estrogen secretion appears to be associated with an increased production of chorionic gonadotropin.

These data would not have meaning as to source unless we consider the placental content of estrogen. The mean value of a large series reported by Diczfalusy is 320 μ g of estrone equivalent per kilogram of placenta. In 10 hours, even after subtracting the first hour's production, our placentas produced an average of 2,700 μ g; so there should be little question about the placenta's ability to elaborate estrogens. Of course, this is only 6 or 7 mg. of estrone equivalent per day, less than 10 per cent of the quantity that Dr. Smith reports in the urine of a pregnant woman at term. I am sure, however, that we have not provided our isolated organ with the same cozy environment that it had in its own womb.

DR. ABRAHAM E. RAKOFF, Philadelphia, Pa.—It is interesting to note that the authors now believe that enzyme hydrolysis followed by acid hydrolysis apparently gives the greatest information regarding urinary estrogen metabolites. I wonder if they have more data concerning the nature of the estriol enhancer for which they have presented evidence. It is well known in the bioassay of estrogens that various combinations of estrogens give rise to results that might not be expected from the simple addition of the 2 estrogens. Is this material that they are speaking of a weak estrogen which is in this same way augmenting the bioassay result? If so, I wonder if they have employed bioassay methods other than the vaginal smear technique, because it is well known that different estrogens in different combination will give different results, depending on whether vaginal smear technique is used or whether, for example, increase in uterine weight due to water retention is used as an end point.

I wonder whether Dr. Smith has used any bile from pregnant women in comparison to urinary excretion in patients to see whether this enhancer was present at the liver level of estrogen metabolism. Some years ago we had the experience of comparing the assay of estrogens in the bile and in the urine of pregnant women. Bile is a very rich source of estrogen, and excretion products in the bile are different from those present in urine. It would be interesting to know whether or not this enhancer substance was already present at this stage.

DR. GEORGE V. SMITH (Closing).—We have been intrigued by the theoretical physiological possibilities of these unknown estrogens. We have wondered whether the assumed push-pull mechanism resulting in ovulation may be related to these unknowns. Perhaps they play a part in the onset of labor and lactation. These ideas are purely speculative but inviting.

Dr. Rakoff asked if we had further knowledge of SP-1. We have not. However, studies thus far of 16-alpha-hydroxyandrostenedione suggest that this intermediary steroid might possibly be SP-1.

We have not used any bioassay method for estrogenic action except the vaginal smears of ovariectomized mature rats. Furthermore, in these studies we have not worked with human bile. In the past we have had great difficulty in the processing of bile for the measurement of its content of estrogens.

EFFECTS OF PREOPERATIVE IRRADIATION ON ADENOCARCINOMA OF THE UTERUS*†

HERBERT E. SCHMITZ, M.D., CHARLES J. SMITH, M.D.,
AND WILLIAM C. FETHERSTON, M.D.,** CHICAGO, ILL.

ENDOMETRIAL carcinoma, by virtue of its intrinsic characteristics that set it apart from cancer of the cervix, allows considerable latitude in acceptable therapeutic regimens. It is not possible, for example, to insist that an established routine of primary surgical treatment submit an apologia to justify a salvage percentage that may be below a critical level of success. The reported 5 year results of contemporary studies utilizing operation or combined therapy are so comparable that differences may be considered to be within the limits of statistical variation. It remains, therefore, on the eve of the ultimate summation of etiological and histopathogenetical knowledge, to delineate the possible avenues of therapeutic approach that are bound to come in the future. In all deference to the practical success of surgical treatment, it must be emphasized that surgical eradication of disease implies a certain degree of failure. Surgery, of its very essence, slashes the Gordian knot of therapeutic confusion but leaves nothing accomplished to further the elemental achievement of basic understanding of a disease process. We hope to point out certain concepts suggested by the results obtained in following a plan of treatment which originally was considered to owe its success to the local control of disease and its effect on tumor environment.

For many years, we have used the combined technique of preoperative radiation followed by operation whenever possible. In any evaluation of this technique, the adequacy of the preoperative radiation must be established if it is to be given credit for improvement in end results. Conversely, failures with inadequate therapy should not be used to discredit this technique. Absence of viable tumor in the surgical specimen after preoperative irradiation has varied from 12.5 to 89 per cent of cases in the various series reported.¹ This discrepancy indicates either differences in preoperative radiation techniques or differences in the criteria for evaluation of persistent viable disease. In contrast to other series, throughout our present study the figure of 75 per cent sterilization of tumor has remained fairly constant. We feel that such a high percentage of sterilization of tumor with our technique is ample justification for continuation of this procedure, particularly in view of the excellent end

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results in this group of cases. The additional advantages accruing to preoperative irradiation, such as damage to persistent tumor and damage to vessels and lymphatic structures, will aid in the prevention of spread of the disease at the time of definitive operation, even where sterilization is not accomplished. Detailed descriptions of radiation changes have been noted in our previous publications.^{2, 3}

The excellent studies on primary operation in the treatment of this disease by Meigs,⁴ Brunschwig,⁵ Douglas,⁶ Javert,^{6, 7} McKelvey,⁸ and others,^{9, 10} have been followed with interest. The marked technical advances in recent decades, both in surgery and in improved radiation techniques, have inevitably improved the end results in this disease. Only continued study and evaluation will ascribe to each modality its proper place.

It is not our purpose to seek a middle-of-the-road compromise between a surgical and a nonsurgical approach. We feel that preoperative irradiation is an adjunct to the therapy of endometrial cancer with an entirely as yet undisclosed salutary influence on the case of *arrested* uterine cancer. By no means do all victims of endometrial carcinoma die by virtue of local recurrence of disease. Factually, in the operable case with reasonable containment of tumor and adequate surgical technique, the hazard of mortality is too often extrapelvic. These cases then are surgical failures, not through technical inadequacy, but by lack of appreciation of the cryptic nature of neoplastic cytemia.

Material

The material consists of a series of 280 cases, half of which have been reported previously.³ A varying degree of selection of cases is inevitable in a series of this type. Some patients are referred following primary operation elsewhere. Some patients refuse preoperative irradiation prior to operation. Some are inoperable at the time of first examination or have recurrent inoperable disease and thus fall into the category of radiation alone. This series will include all cases admitted to the Gynecologic Service of the Mercy Hospital Institute of Radiation Therapy, regardless of extent of disease or previous therapy.

For comparative purposes, the cases are divided into a series prior to 1946, which has been previously reported,³ and a series from 1946 to 1953, inclusive, in which more precise and refined techniques were applicable, and for which 5 year statistics are available. As is seen in Fig. 1, the over-all uncorrected 5 year survival has risen from 47.4 per cent in the original series prior to 1946 to 67 per cent in the group treated since 1946. Thus, the over-all survival rate in this unselected group has improved 20 per cent and this is attributable to improvement in both radiation and surgical techniques.

Plan of Treatment

In the present series from 1946 to 1953, 71 patients received treatment according to our preferred techniques. This consists of the insertion of the Y radium applicator on 3 separate occasions for a dose of 2,000 mg. element hours per application, or a combined total dose of intracavitary radiation of 6,000 mg. hours. The loading, utilization, and isodose curves for the Y applicator have been previously reported.^{11, 12} These applications are at weekly intervals, with the administration on intervening days of deep x-ray therapy through

2 ports, anterior and posterior, with a 1,000 kv. machine and adequate stripping in the midline to protect bladder and rectum from overdosage. Following the third intracavitary application, daily external irradiation is continued until a total dose at the midpelvis of 3,500 to 4,000 r is administered. The actual dose to the endometrial surface from the intracavitary radiation is quite high—of the order of 20,000 to 40,000 gamma r.

An adequate dosage is delivered to the usual depth of the myometrium but some variation at depth is inevitable in the grossly distorted myomatous uterus. The contribution from external therapy, added to the intracavitary radiation, produces close to 5,000 r to the lateral pelvic fields, so that all potentially tumor-bearing areas in the pelvis receive what is generally considered to be an average cancericidal dose of radiation. That this so-called cancericidal figure will vary from tumor to tumor is well recognized, but this type of planned attack achieves maximum radiation benefits in any case.

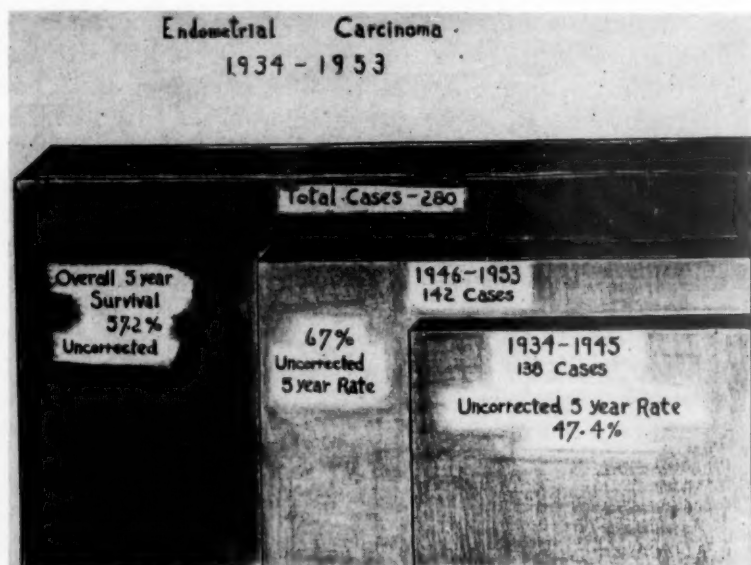


Fig. 1.

The above program must, of course, be varied at times depending upon the clinical problem presenting. In the markedly obese patient, for example, even with supervoltage therapy, it is at times difficult to deliver 3,500 to 4,000 r to the midpelvis through 2 portals and more portals may be utilized. In certain cases, the intracavitary dose may be increased and the external therapy reduced. In general, the plan is to deliver as close to the above dose as commensurate with safety.

This program is followed by operation in 6 to 8 weeks. At least total abdominal hysterectomy, with a generous cuff of vagina and bilateral salpingo-oophorectomy, is performed. No effort is made to suture or otherwise tamponade the cervix. Tissues are handled gently and major vascular and lymphatic channels are ligated and severed as early in the procedure as is feasible. On occasion pelvic lymphadenectomy and parametrial excision out to the lateral wall are performed, depending upon the age and the physical condition of the patient and the apparent extent of the lesion. This procedure is particularly applicable in cases of cervical extension of the disease, for in these cases extension and metastases frequently followed the pattern of cervical carcinoma, as has been pointed out so well by Heyman,¹³ Kottmeier,¹⁴ and others.

Much of the plan of surgical attack is dictated by the findings at the initial curettage and pelvic examination under anesthesia. The importance of this initial examination by curette and manual palpation cannot be overemphasized.

Radiation Changes

All surgical specimens are sectioned according to a technique developed in preparation of a previous report. Of the 71 cases treated with preoperative radiation followed by operation, 55 showed no tumor in the surgical specimen—a sterilization rate of 77.4 per cent. Similar studies on specimens in the group prior to 1946 showed a sterilization rate of 57 per cent. This increasing effectiveness in eradicating viable tumor with preoperative radiation we ascribe to improvement in techniques. This makes it possible to deliver a higher mid-pelvic dose of radiation with safety. As Koletsky¹⁵ points out, "The injury

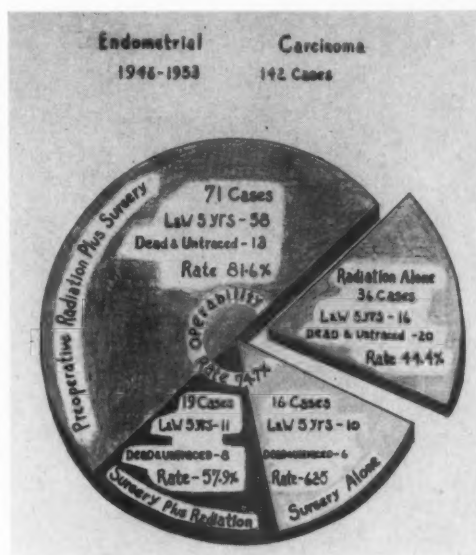


Fig. 2.

of (neoplastic) tissue from external sources (x-rays, gamma rays, and fast neutrons) depends...in degree...on the type of radiation, its energy and rate of delivery, the total dose and also the portions of the body exposed. . . . All forms of ionizing radiation have qualitatively similar effects. These effects usually become apparent only after a latent period which varies from a few minutes to years." Radiation changes in non-tumor-bearing tissues were described in detail in previous publications.^{2, 3} These consist in brief of radio-necrotic plaques within the cavity of the uterus and vascular and tissue changes, characteristic of radiation reaction. We have demonstrated the typical destructive effects of ionizing irradiation on adenocarcinoma of the fundus with typical changes in the nucleus and cytoplasm of irradiated cells. The end stage is hyalinization.

Results

Material studied consisted of 280 cases in which there was an over-all 5 year survival rate of 57.2 per cent. This material for comparative purposes is divided into 2 chronological groups. The first series of 138 cases treated between 1934 and 1945, inclusive, showed an over-all 5 year survival rate of 47.4

per cent. The second group of 142 cases treated from 1946 to 1953, inclusive, showed an over-all 5 year survival rate of 67 per cent. This improvement of 20 per cent in the over-all figures can be attributed to improvements in all phases of therapy, inasmuch as it includes cases treated by all modalities. Of the 142 cases in the present series, 106 cases were operated upon, giving an operability rate of 74.7 per cent. This operability rate is rising in our own more recent cases, as well as in other reported series. This will inevitably lead to an even greater salvage.

The most significant improvement, as can be seen from Figs. 2 and 3, occurred in the group of the 71 cases treated by preoperative irradiation followed by operation. In this group of 71 cases, the over-all 5 year survival was 58

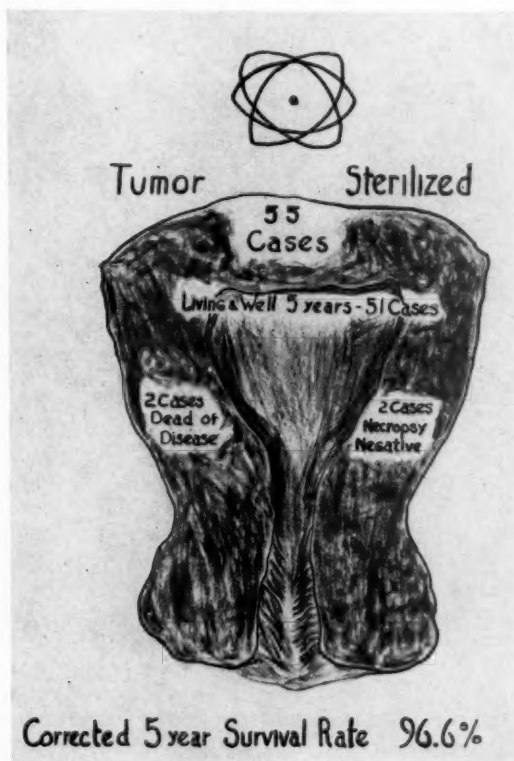


Fig. 3.

cases or 81.6 per cent. If we consider by further refinement the cases so treated which showed no viable tumor in the surgical specimen, we find 55 cases with 4 deaths. Of these 4 deaths, one patient died 2 years postoperatively of mesenteric thrombosis, and no evidence of tumor was present at autopsy. A second died of coronary thrombosis 2 years following operation and no tumor was found at autopsy. The two remaining patients died of recurrent or metastatic disease. Deducting the 2 with no disease at autopsy, there were 53 patients with no tumor present in the surgical specimen, with the corrected 5 year survival in this selected group being 96.6 per cent.

Comment

In addition to tumor destruction in this large percentage of cases, it is our contention that the radiation effect, even in the presence of persistent tumor,

retards the spread of tumor cells. Apparently viable tumor cells have been demonstrated in the peripheral blood at the time of operation for neoplastic disease by Cole¹⁶ and others.¹⁷⁻²¹ Various cytotoxic agents have been employed following operation in certain malignancies in an effort to combat this phenomenon. It has been our feeling that even where tumor persists after radiation the cells which might be spread at the time of operation would be so damaged as to make their survival unlikely.

Neoplastic cytemia has been subject to consideration since 1869.²² It has not been until recently, however, that the pharmacological countermeasures have been instituted. We submit, moreover, that there are other aspects to the problem than bringing the circulating tumor cells into contact with some cytotoxic agent. Engell¹⁷ has claimed little relationship to exist between manipulation and the number of patients who demonstrate circulating tumor cells. Too often we overlook the biological truism that invasion or dissemination can occur only in a susceptible host. While it is true that this may be only a matter of degree, nevertheless, the longevity of some hopelessly involved patients cannot be otherwise explained. Green²³ has demonstrated experimentally that there is variation in the virulence of tumor transplants, but what we cannot ascertain is whether he may not have demonstrated a variation in antigenic stimulation between tumors. Smith and co-workers²⁴ have proposed that the presence of noxious stimuli provokes a somatic response which can be detected as basal cell activity. Scott²⁵ has gone beyond this to suggest that the interplay between cancer and tissue resistance depends upon the availability and utilization of iodine and the enzymatic destruction of mast cells to liberate cytotropic hormones, resulting in uncontrolled growth. Furth²⁶ states that neoplasia can arise (1) by sustained deficiency of the restraining factors (of cell growth), (2) by sustained excess of the stimulating factors, or (3) by altered responsiveness of either the target organ or the regulators—in other words, a derangement of the feed-back mechanism of cellular growth control.

It is conceivable, then, that irradiation provides a twofold antineoplastic effect, namely, the specific metabolic inhibiting effect through nucleus injury, and what might be termed a general deterrent effect by raising the host resistance either through a general somatic response to a profound physiological injury or by the increase in body resistance mechanisms through the stimulation of radiant energy. A case in point is that of Mrs. H. C., who showed no clinical evidence of recurrence of breast carcinoma despite a progressive cachexia. Bone marrow aspiration on 5 different occasions demonstrated the peripheral hematopoietic system to have undergone almost total replacement by tumor cells. After treatment with x-ray, oophorectomy, and cortisone, subsequent bone marrow aspirations revealed only fibrous tissue with marked clinical improvement in the patient. This suggests that not only can the host resistance be varied, but the neoplastic process can be subjected to some degree of reversal. Fractionated irradiation over a 4 to 5 week period would be suitably adapted to develop this individual tumor defense, and this lapse of time, rather than being the liability so often alluded to, actually serves the patient's interests to a greater extent than we hitherto considered.

Conclusions

1. Preoperative irradiation by this technique in the operable group of patients will destroy endometrial carcinoma in 75 per cent of cases and has produced an 81.6 per cent 5 year survival rate.
2. It is possible to salvage 96 per cent of patients so treated and found to have no residual tumor.

3. Combined preoperative irradiation and operation have produced a greater salvage rate than any other method in this series.
4. A 20 per cent improvement in 5 year end results coincided with the installation of improved radiation facilities.
5. A concept of improvement in host resistance to tumor dissemination is submitted

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Discussion

DR. SAUL B. GUSBERG, New York, N. Y.—Dr. Schmitz has produced an enviable record of 76 per cent sterilization of tumor in the surgical specimens, an 81.6 per cent 5 year survival in patients treated by this combined mode, with a net corrected cure rate of 96.6 per cent for those without residual tumor at hysterectomy. Concerning the meticulous nature of his dispersal technique of preoperative radium application and its adjuvant x-ray therapy, or the efficiency of the surgical treatment following this, there can be no question. I could ask some other questions concerning the possible use of a packing method of radium application or a supplementary application to the vaginal vault; I could suggest that x-ray therapy might possibly be postponed to the postoperative period; but these variations are insignificant compared to my agreement with the basic principles of this work: first, that individual tumor biology is a most important consideration in corpus cancer for prognosis and treatment, and may indeed, I believe, be an index of the host's resistance, and, second, that radiation can have a profound and beneficial effect on these tumors and the patients bearing them.

Yet, the very efficiency of the combined mode of therapy together with its close competition in excellence of result by primary operation alone force one to the conclusion that there must be factors of selection that could be exploited in a clinical staging that would benefit the codification of treatment of this disease as much as has that of cancer of the

cervix. I think these factors are, first, the size of the uterus, which is still a fairly good index of local growth in a tumor whose tendency is to confine itself to the uterus so commonly; second, lack of tumor differentiation, which offers another clue to myometrial penetration and permeation of pelvic lymphatic channels; and third, involvement of the cervix, which will open another lymphatic pathway that must be encompassed surgically or radiotherapeutically. These factors can be appreciated at curettage and examination under anesthesia to make a clinical staging that will be meaningful for treatment.

If one applies such a staging to the material of our clinic it seems clear that the small, well-differentiated tumors can be treated by hysterectomy only; the tumors of moderate extent will benefit strikingly from combined therapy, whereas those with deep myometrial penetration and/or cervical involvement may require more radical radiation or more radical operation.

DR. FRANKLIN L. PAYNE, Philadelphia, Pa.—During 25 years Dr. Schmitz has accumulated and documented sufficient material to be of real significance: 138 patients in the first series and 142 in the last. Comparison reveals that his radiation sterilization rate rose from 57 per cent in the first to 77 per cent in the last series, while the survival rate rose from 47 to 67 per cent, respectively. He chose for special comment 71 instances of the ideal treatment, with a survival rate of 82 per cent. Among the 55 for whom preoperative radiation sterilization was achieved, the uncorrected survival rate rose to 91 per cent and if 2 unrelated deaths are deleted this figure reaches 96 per cent.

The reasons for this improvement in therapeutic accomplishment seem to fall into three groups. First is the increased radiation effectiveness in eradicating viable tumor, damaging persistent tumor, and rendering blood vessels and lymphatics less capable of malignant cell transportation. Second is the rise in operability to 75 per cent. No doubt this is due to both improved material and the inclusion in the combined therapy group of many patients who formerly would have received radiation alone. A comparison of the 44 per cent radiation survival rate to that of 82 per cent following combined therapy demonstrates the justification of this calculated risk. Here a point of difference arises. Dr. Schmitz says, "Much of the plan of surgical attack is dictated by the findings at the initial curettage and pelvic examination under anesthesia." With both cancer of the cervix and of the corpus, we do at this time indicate the clinical classification and the proposed management, but for the latter (cancer of the corpus) the original plan should not be immune to revision. Experience indicates that re-evaluation, 4 to 6 weeks following the termination of the radiation or even later at times, frequently permits elevation from the level of further radiation therapy to that of the combined approach with its attending increase in life expectancy.

The third reason for improved results concerns neoplastic cytemia and host resistance. We are in agreement with Dr. Schmitz that, following adequate radiation therapy, the errant tumor cells may well be so damaged as not to survive in distant areas. The term "adequate" should be emphasized for mere token or ineptly applied radiation is worse than no therapy because of the false sense of security that it gives the surgeon at the time of operation. Recent reports upon neoplastic cytemia have brought home the importance of this consideration, which applies even more forcibly to primary operation for cancer of the corpus. The primary surgeon should not ignore its implications, nor can he depend upon the intravenous chemotherapeutic agents to vitiate the potential of induced peripheral neoplastic cell showers—in the light of the recent reports by Cole and by the Roswell Park group.

The question of host resistance to malignant cells is really the crux of the cancer problem, as to both local spread and distant metastasis. Much has been accomplished in the determination and the enhancement of immunologic response to infectious disease—but not so with neoplastic disease. In the former field, both clinical observation and immunologic studies suggest strongly that radiation reduces the immunologic response to the infectious process. With neoplastic disease, the exact nature of the host resistance has not been determined as yet. Thus far no good experimental evidence has been produced to indicate that radiation in the therapeutic dose range either increases or decreases this resistance.

The more refined immunologic techniques may now permit investigators to throw light on this question and to explain Dr. Schmitz's unequalled clinical accomplishment in the management of cancer of the corpus.

DR. AXEL N. ARNESON, St. Louis, Mo.—The statement often made by our well-remembered Emil Novak that, "the treatment of endometrial cancer remains in a fluid state," is descriptive of the situation here. The ebb and flow of opinion for operation alone or for combined treatment with radiation produces a debate that keeps alive the attempt to explain why preoperative irradiation may affect favorably the end result of treatment. Attention has been focused primarily on the local area. The essential factors assessed in relation to prognosis include uterine size, histology of the tumor, and persistence or disappearance of cancer within the uterus after preoperative treatment. Proponents of the combined treatment argue for each of these factors certain physical and biological data that support the use of preoperative irradiation. The crux of those arguments involves the rather poorly understood factors of host-tumor relationship. Now, for the first time, Dr. Schmitz extends investigation beyond the local area by considering the regular habit of tumor cells to appear in the peripheral circulation and in the bone marrow. I believe that is important for the better understanding of so-called "host resistance."

I cannot let pass this opportunity to compare Dr. Schmitz and his 2 co-authors with the 3 princes of Serendip. While engaged in a search for riches they came upon other discoveries of greater value. From that experience Walpole coined the term "serendipity." This does not, in my opinion, imply an accident. If one is not looking he does not come upon anything to see. It is not unlikely that the irradiation of tissues produces changes, perhaps chemical, and certainly subtle, that give rise to constitutional effects of biological significance in the control of cancer.

DR. JOHN L. MCKELVEY, Minneapolis, Minn.—Dr. Schmitz has presented a most persuasive paper. It is one of the few papers dealing with this subject which is acceptable from a statistical point of view. His paper reports a control series of cases expressed *in toto* and as compared with similar material from the same clinic but handled in a somewhat different way. That is the only method by which one can arrive at an acceptable statistical expression of the problem.

The question is, have we seen all the story? Unfortunately I have not seen the paper which Dr. Schmitz summarized. Dr. Gusberg has seen the paper and has stated that in these 2 series there has been one significant change in addition to the change in radiation technique involved. I would like to ask Dr. Schmitz to state for us what has been the change in the surgical rate as between these two groups.

I will not bore you with the details of our study. It has already been presented under a variety of circumstances. By reversing this process, we withdrew irradiation and increased the surgical rate, and, as we did, we increased the survival rate. I am talking only about routine application of radiation. Irradiation does have a part to play in specific problems. We can find no effect in our material of the addition of radiation to the surgical attack. Indeed there is little doubt in the minds of most of us but that the basic feature is the surgical rate which is applied and that this controls, for practical purposes, the survival rates. The question is, have we exhausted the possibilities for surgical attack? Almost every woman with endometrial carcinoma can be treated surgically for this disease. We have achieved a survival rate which apparently represents the survival of almost all patients who have localized tumor within the uterus. The problem now is to search for more effective methods of handling extended tumor.

DR. LUDWIG A. EMGE, San Francisco, Calif.—Dr. Schmitz's excellent analysis brings up several problems well worth discussing, among them the still unanswered question of how to select the most promising method of therapy for endometrial cancer. That preoperative irradiation is a "must" has been repeatedly claimed on the basis of well-substantiated figures and, again, finds support in Dr. Schmitz's analysis. On the other hand, valid claims have been made that proper selection of cases obviates this procedure and thereby speeds up the therapeutic approach without necessarily jeopardizing the chances for a

cure. My own experience has taught me that neither side necessarily is correct but that the principles involved depend upon astute clinical selection of therapeutic measures best suited to the individual.

Over a period of 20 years (1930-1942 and 1946-1954), I have evaluated and treated 124 private patients afflicted with endometrial cancer. Four were lost to follow-up during the war years. An analysis of 120 patients based on an accurate follow-up is presented in Table I.

TABLE I. SURVIVAL STUDY OF 120 PRIVATE PATIENTS TREATED FOR ENDOMETRIAL CANCER

THERAPY	TOTAL NO.	DIED UNDER 5 YEARS	SURVIVAL IN YEARS			
			5 PLUS	6-10	11-15	16-20 PLUS
Operation only	58	2 (3.4%)	56 (96.6%)	49 (84.5%)	22 (37.9%)	16 (27.6%)
Radium and operation	27	6 (22.2%)	21 (77.8%)	19 (70.4%)	10 (37 %)	6 (22.2%)
Operation and postoperative x-ray	17	9 (52.9%)	8 (47.1%)	6 (35.3%)	5 (29.6%)	2 (11.7%)
Irradiation only	16	7 (43.7%)	9 (56.3%)	7 (43.7%)	3 (18.7%)	2 (12.5%)
Not treated	2	2 (100 %)				
Total	120	26 (21.7%)	94 (78.3%)	81 (67.5%)	40 (33.3%)	26 (21.7%)

The material is divided into five groups listing the survival incidence in total figures and percentages. It is evident that in this series the 5 and 10 year survival rates for operation alone are consistently greater than those for irradiation and operation combined. I believe this to be attributable to the proper evaluation of the individual cancer problem. My approach to the problem has been as follows:

1. Patients past the menopause with a highly differentiated carcinoma, as determined by endometrial office biopsy, with a cytologically determined low estrogen level, a freely movable uterus, and no detectable evidence of metastatic spread were chosen for primary operation.

2. The selection of the second group was based on age levels prior to and including the menopausal years, the discovery of a poorly differentiated and actively growing type of cancer, the cytologic evidence of an active estrogen function, and palpatory evidence of contour changes in the reproductive organs.

It must be acknowledged that the two groups are biometrically dissimilar and that the decided difference in results therefore may have, in part, been due to chance. Time does not permit a discussion of the causes of death among those having lived longer than 5 years but cancer was relatively uncommon in Group I.

3. Group III originally was selected on the same basis as Group I but findings at operation, or at the subsequent tissue study, proved the selection to have been in error. Although irradiation was promptly instituted I have doubts that it materially improved what had been accomplished by radical operation in this group.

4. Groups IV and V incorporate those patients whose physical state contraindicated surgical procedures or in whom the extent of malignant dissemination held out no hope for amelioration.

The combined survival rates bring the 5 year survival incidence for this series to 78.3 per cent and the 10 year rate to 67.5 per cent, figures which compare favorably with the best therapeutic results so far reported. These figures are offered in support of my conviction that the individualization of therapeutic measures in the treatment of cancer in general and endometrial cancer in particular leads to a higher return in terms of survival than the unconditional acceptance of a single standard method of therapy.

DR. JOHN B. MONTGOMERY, Philadelphia, Pa.—Dr. Schmitz has presented a paper which tends to support a plan of therapy to which we have subscribed in our clinic for many years. Several years ago Dr. Scheffey presented a brief series of patients treated with

multiple intrauterine capsules with a 5 year survival. We have now about 136 patients treated prior to 1953 with preliminary irradiation according to a planned method in which the radium was placed in the endometrial cavity at the time of the initial diagnostic curettage. The pathologist's report was available within 6 hours. In 58 per cent of the cases no residual carcinoma was found on routine examination of the removed uterus. Among this group, the 5 year survival rate was 93 per cent. When residual malignancy was found the 5 year survival fell to 68 per cent.

DR. NORMAN F. MILLER, Ann Arbor, Mich.—I would raise the question, are we not focusing too little attention on an area even as important as therapy? It is obvious that one may cure selected groups of cases of endometrial carcinoma by either operation or irradiation or by a combination of both.

It is undesirable to leave the impression to readers of the future that either operation or operation and irradiation is the most important thing about endometrial carcinoma. Other aspects are also important.

We should recognize that there is no satisfactory way of clinically staging endometrial carcinoma. The sooner we recognize this the sooner we come to grips with the need to devise other methods, if possible, for comparison.

Also, we must recognize that we are not doing very well with endometrial carcinoma. If one considers all cases, not selected groups, results are not so good.

In our 544 cases, carcinomatous metastasis to the lung was the most common finding. This conclusion is supported by the work of Henriksen and by a recent report by Bunker.

To arrive at a diagnosis, we curette the uterus and do a thorough job. In doing this we may send a shower of healthy tumor cells into the blood stream. Where do they go? Into the vessels of the pelvis, on into the inferior vena cava to the right side of the heart, and on into the lung. Some cells may go through the arteriovenous shunts into other parts of the body. Maybe this means that it is important to find other better ways of making a diagnosis.

Finally we should consider the question of why, in menopausal women, we have hyperplasia, anaplasia, and neoplasia. In the long run, prevention may prove more fruitful than attempts at cure.

DR. SCHMITZ (Closing).—We must agree with Dr. Payne's remarks as to the extension of surgical dissection and improvement of surgical technique. That is happening in all published series. That is what I have told Dr. McKelvey on the occasions when we have had the opportunity to present two sides of the question—that his surgical technique has improved. When you go back and compare the results that were obtained by your predecessor or by yourself 10 years ago, in all probability you are guilty of selecting and doing an injustice in your results.

The packing method, Dr. Gusberg, has been proved to be the most successful. It gives a homogeneous radiation to the endometrial surface of the uterus. It also carries with it the highest number of complications and, as we do not have such highly skilled men in our clinic in Chicago, we stay with the simpler method of the Y capsule, which is second in line of satisfaction.

In our first report we grouped our patient material not only clinically but also microscopically. We wanted to see whether or not there was a difference between Grade I and Grade IV. Apparently there was none because, if you have intense enough irradiation, you can destroy anything, including normal tissue, and we can use a sufficient strength of irradiation to destroy the adenoma malignam as well as the anaplastic tumor. Clinically, we used a grouping very similar to yours. We may have gotten it from you, but it has been used in our clinic and when anyone asks me to define what is meant by Group I cases clinically, I have great difficulty. I talk about size and where I found the tumor with my curette, but there is nothing definite about it and I do not think it enters into the situation here.

We bumped into these considerations of tumor cells in the circulation in the second study. We did not think about this, even though the knowledge of dissemination of cells in the circulation was available, but when the chemotherapists began to study the effect of

various agents on tumor cells, we realized that this was a major problem. We could continue our study and by collecting tumor cells that are loose in the circulation we can study the effects of radiation on those cells. I am sure that the antigenetic effect and other effects of radiation which may be secondary take some time to occur, and maybe that is why there is a benefit to the 6 to 10 week interval or what McKelvey calls the delay period. That may be a good thing for the patient. Those cells are so sick that when they get into the circulation, with the possibility of improved host resistance due to therapy, that may be the thing that improves our results.

Dr. Emge and Dr. Gusberg have excellent results. If one surgeon with surgical ability and surgical technique can raise his statistics on survival by using preoperative irradiation, then I think we have something to offer. Dr. Miller uses x-ray alone as a general pattern. If I had to decide between the 2, I would choose x-ray alone because I have been looking for secondary effect on the cell. But when someone says, "We used radium" that is just like my saying, "We operated." How did we operate? What did we accomplish? We have to be more definite. Take a series of cases and study it. For instance, McKelvey has a 2 year series with not a Group IV case in the list; that makes a difference too. If you have an over-all sampling of material and you account for all of it, not some of it that stayed in the admitting clinic but all of them that approached the door for admission, then the figures mean something entirely different.

Let me say this to Dr. Miller: I am sure we agree as far as selectivity of groups of carcinoma patients. Unless you take an over-all sample, the increased survival rate so far as irradiation or operation is concerned is going to be dented because of another factor that enters into this: we are seeing these carcinomas and are recognizing them ourselves at an earlier date.

THE RELATIONSHIP OF THE SQUAMOCOLUMNAR JUNCTION AND THE ENDOCERVICAL GLANDS TO THE SITE OF ORIGIN OF CARCINOMA OF THE CERVIX*

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THE problems of neoplastic disease occupy an increasing share of the physician's time and attention. The modern concept of therapy stresses that treatment will more likely be successful if the neoplastic process is detected and treated in the early and localized stage of its development. This concept will remain true until more effective methods of therapy are available than operative extirpation, irradiation, and present-day chemotherapeutic agents. This concept is especially exemplified by the generally accepted attitude toward carcinoma in situ of the uterine cervix. This subject was probably first discussed in the American literature in 1910 by Rubin,¹¹ who pointed out the significance of complete pathologic examination and suggested that the diagnosis of carcinoma of the cervix was possible in its preclinical stage on the basis of the intrinsic morphology of the epithelial cells. These morphologic changes in surface epithelium were recognized in 1922 in a case reported by Martzloff.⁶ Subsequently he was able to demonstrate lymphatic invasion in this specimen after serial sections of the entire cervix were made.

This study is concerned with the relationship of the squamocolumnar junction, or transformation zone, and the types of epithelia of the uterine cervix involved in the development of carcinoma in situ and invasive cancer. Squamous cell carcinoma of the cervix is expected in the portio vaginalis, normally covered by squamous epithelium. However, the neoplastic process also arises in the endocervix from the epithelium lining the endocervical canal and the endocervical glands. The anatomic location of the transformation zone is of importance in the detection of early stages of cervical malignancy. Foote and Stewart⁴ have emphasized the importance of this zone in obtaining punch biopsy specimens for the diagnosis and distribution of carcinoma in situ. The material to be presented will demonstrate the marked anatomic variation in the location of this zone and variations in the relationship of the epithelia encountered in and about this area.

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Meyer^{7, 8} in 1910 discussed his concept of the development and changes of the epithelium of the portio and the cervix. This zone of different types of epithelia would not appear to be static. The histologic picture is made more complex by the morphologic potential of the small, undifferentiated basal cells, or reserve cells, lying beneath the mucous epithelium of the endocervix and endocervical glands. These reserve cells are important because they have a potential to undergo hyperplasia and to differentiate to squamous epithelium, as pointed out by Carmichael and Jeaffreson.^{1, 2} The studies of Howard, Erickson, and Stoddard⁵ support this concept, and, as shown in their illustrations, these hyperplastic basal cells may be delineated from portio squamous epithelium.

Material

This study is based upon the histologic findings in 200 cold-knife cone specimens of the cervix obtained for the investigation of suspicious or positive exfoliative cytologic spreads, or for more definitive study of cervixes showing carcinoma in situ or severe dysplastic change in punch biopsy specimens. The smallest cone specimen measured 1.0 cm. across its base and 1.0 cm. in length. The atrophic cervix of the postmenopausal patient frequently limited the size of the specimen obtained. The largest specimen measured 3.0 cm. across its base and 3.5 cm. in length, and three of these specimens showed endometrial tissue in the apex of the cone. The average length of the cone, on the basis of length of the endocervix, was 2.5 cm., and the diameter of the base was 2.0 cm. The minimum number of sections studied per cone was 6 and the maximum was 107, with an average of 16 sections per cone specimen. This study is subject to the valid criticism that a complete histologic picture may be obtained only by serial section of the specimens. The number of cone specimens involved in the study makes this type of definitive investigation somewhat impractical. It was our feeling that the study of this number of specimens by the method described would at least permit the suggestion of the validity of certain observations.

Transformation Zone

A meticulous study of 697 uteri and 156 cervixes by Schneppenheimer, Hamperl, Kaufmann, and Ober¹³ points out the variability of the anatomic location of the squamocolumnar junction, and they suggested that the relationship of the epithelia in this area appeared to be related to age and ovarian function. They found the squamocolumnar junction to be located at the clinically apparent external os in only one specimen. In their manuscript they have sketched the 9 types of transformation zones which might be expected. Not having observed true erosion in any of the 853 specimens studied, they classified their material under six groups according to the observed variations in the squamocolumnar junction.

The material of this study has been divided into five groups. In 172 specimens, the squamocolumnar junction could be determined and these cones are classified under four groups as shown in the sketches in Fig. 1. It was not possible to plot the squamocolumnar junction in 28 specimens because of: (a) previous multiple punch biopsies, (b) previous cauterization of the cervix prior to adequate investigation, (c) faulty technique in obtaining the cone specimen, or (d) poor technique in preparing the sections in the pathology laboratory.

The numbers of specimens classified under the various types are shown in Table I. There would appear to be no significant difference in the average age of the patients classified as Type I, Type II, or Type III. There were 6 nulliparous patients included in the first classification, one in the second, and 2 in the third. During the reproductive years the cervical glands often extend to the portio vaginalis and either communicate freely with the surface or are overgrown by squamous epithelium, as illustrated in Figs. 1 and 2. The average age of patients included under Type IV was 51.4 years, and the endocervical glands in this group were restricted to the endocervical canal and the squamous epithelium extended into the canal. There were no nulliparous patients in this small group.

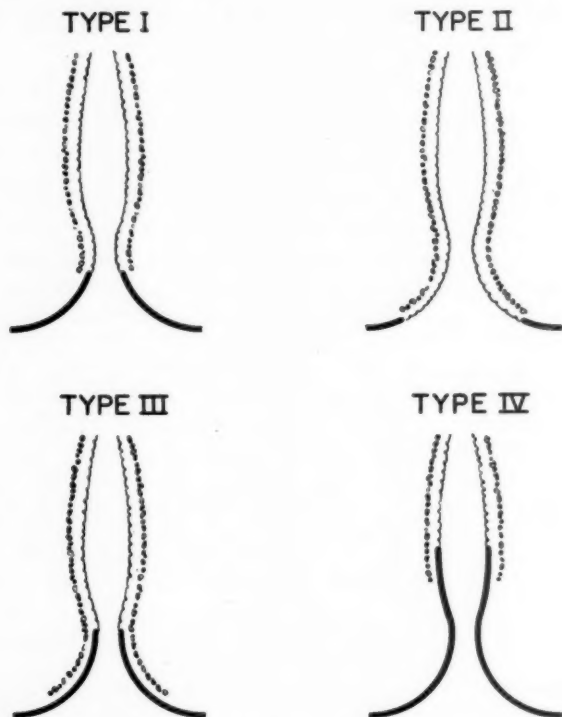


Fig. 1.—Schematic sketches of the four types of squamocolumnar junctions encountered in the study of the cold-knife cone specimens of the uterine cervix. Heavy black line indicates squamous epithelium. Serrated line denotes columnar epithelium of endocervix, and small circles represent endocervical glands. The endocervical glands may be found beneath the squamous epithelium as shown in Types III and IV. The columnar epithelium may extend out on the portio (ectropion) as shown in Type II.

Location of Lesion

An attempt was made to determine histologically the location of the lesions for which the conizations were done in relationship to the squamocolumnar junction and to determine if the process began in the columnar epithelium of the endocervix, in the epithelium of the endocervical glands, or in the squamous epithelium of the portio vaginalis. The neoplastic potential of the basal cells lying beneath the columnar epithelium has been discussed by others.^{1, 2, 5, 12} This potential is portrayed in Tables II, III, IV, and V. Of the 172 specimens in which it was possible to plot the transformation zone, the portio squamous epithelium was involved in only 6 instances. This

type of epithelium seemed to be the most advanced site of the lesion in only one of the 6 instances in which portio epithelium was involved. Of the 28 cones in which it was not possible to plot the transformation zone, the squamous epithelium was involved in one instance, as shown in Table VI.

TABLE I. NUMBER OF SPECIMENS IN EACH TYPE WITH RESPECT TO AGE

TYPE	YOUNGEST (YEARS)	OLDEST (YEARS)	AVERAGE (YEARS)	NO. OF CONES
I	19	69	40.3	40
II	22	59	36.6	24
III	19	79	36.7	101
IV	31	70	51.4	7
†	23	69	43.9	28
Total				200

TABLE II. TYPE I. JUNCTION—SITE OF LESION*

Endocervical epithelium	12
Glandular epithelium	7
Endocervical and glandular epithelia	20
Squamous epithelium	1
Squamous and endocervical epithelia	0
Total	40

*All except three lesions near junction.

TABLE III. TYPE II. JUNCTION—SITE OF LESION*

Endocervical epithelium	5
Glandular epithelium	1
Endocervical and glandular epithelia	17
Squamous epithelium	0
Squamous and endocervical epithelia	1
Total	24

*All except two lesions near junction.

TABLE IV. TYPE III. JUNCTION—SITE OF LESION*

Endocervical epithelium	21
Glandular epithelium	9
Endocervical and glandular epithelia	67
Squamous epithelium	0
Squamous and endocervical epithelia	4
Total	101

*All except five lesions near junction.

TABLE V. TYPE IV. JUNCTION—SITE OF LESION*

Endocervical epithelium	3
Glandular epithelium	2
Endocervical and glandular epithelia	2
Squamous epithelium	0
Squamous and endocervical epithelia	0
Total	7

*All except one lesion near junction.

The lesion was found in the region of the transformation zone in 161 of the 172 cones in which it was possible to identify the zone on histologic sections. In 11 instances, not including the 28 specimens shown in Table VI, the lesions appeared at a great distance from the transformation zone. These

findings are in agreement with the panoramic studies of 100 cervixes by Przybora and Plutowa.¹⁰ Their studies revealed the neoplastic process to be limited to the portio in only 3 specimens.

TABLE VI. TYPE OF JUNCTION UNDETERMINED—SITE OF LESION

Endocervical epithelium	3
Glandular epithelium	0
Endocervical and glandular epithelia	24
Squamous epithelium	0
Squamous and endocervical epithelia	1
Total	28

The incidence of involvement of the various types of epithelia in the 200 cone specimens is shown in Table VII. The surface epithelium of the



Fig. 2.—Endocervical gland communicating with surface of squamous epithelium.

endocervix was the only site of the lesion in 44 specimens, and only the glandular epithelium was involved in 19 instances. In addition to the 19 instances in which only the glandular epithelium was involved, there were 130 instances in which both the glandular and endocervical epithelia were involved. Przybora and Plutowa¹⁰ state that in only 13 of the 100 cervixes examined by them was the cancerous epithelium found exclusively on the surface without

TABLE VII. INCIDENCE OF THE INVOLVEMENT OF THE VARIOUS TYPES OF EPITHELIA

Endocervical epithelium	44
Glandular epithelium	19
Endocervical and glandular epithelia	130
Squamous epithelium	1
Squamous and endocervical epithelia	6
Total	200

extension to the endocervical glands. They found more or less abundant and deep penetration of abnormal epithelium into the endocervical glands in the remainder of the specimens examined by them. Fennell³ found the glandular epithelium to be involved in 93 per cent of the 118 specimens examined by

him. He found the endocervix to be involved in all specimens, and in only 25 did the disease extend on to the portio or non-gland-bearing portion of the cervix.

Types of Lesions Encountered

The final classification of the lesions encountered in the 200 cold-knife specimens is shown in Table VIII. We believe the diagnosis of carcinoma in

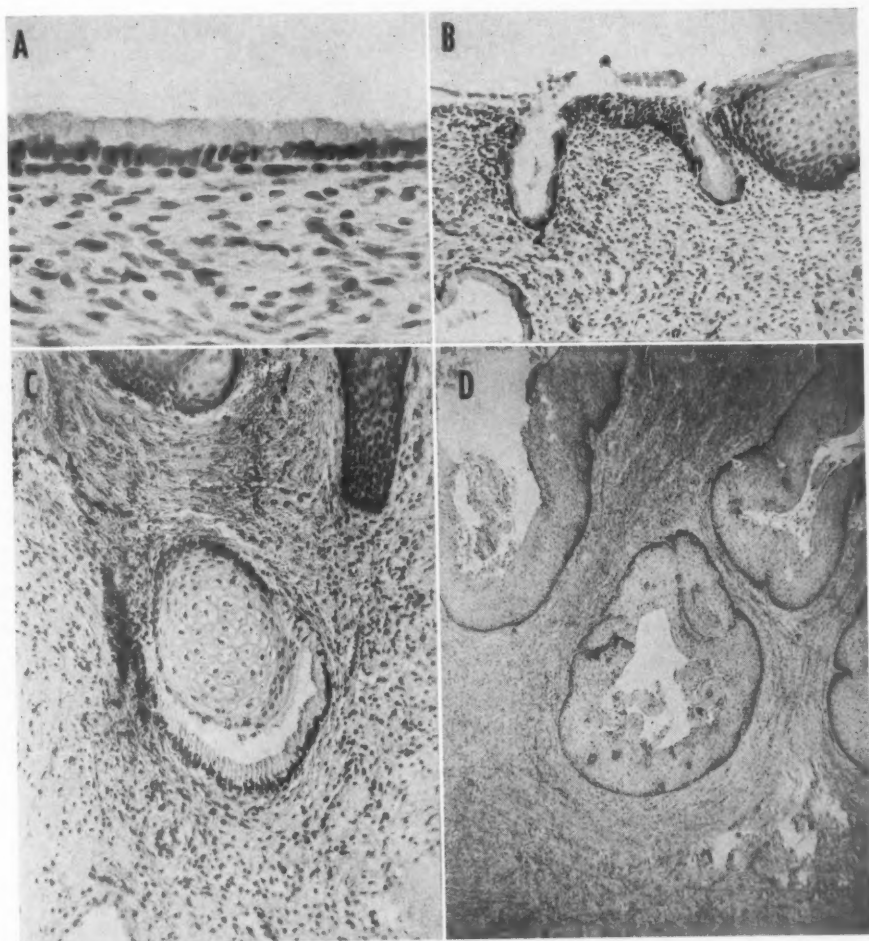


Fig. 3.—A, A single layer of basal, or reserve, cells underlying columnar cells of the endocervical epithelium. ($\times 760$; reduced $\frac{1}{2}$.) B, Hyperplasia of reserve cells with endocervical glands on each side. ($\times 95$; reduced $\frac{1}{2}$.) C, Squamous metaplasia of endocervical glandular epithelium. ($\times 200$; reduced $\frac{1}{2}$.) D, Example of squamous metaplasia of glandular epithelium with pseudopapillary formation. ($\times 42$; reduced $\frac{1}{2}$.)

situ of the cervix uteri has been conservative in our Department of Pathology. At times it was difficult to decide whether the process represented severe dysplasia or carcinoma in situ. It should be emphasized that all of these specimens were studied in pathology conference, and although opinions were not always unanimous, in no instance did the diagnosis established represent the opinion of only one individual. Invasive cancer was not clinically suspected in any of the 18 patients in which these changes were histologically demonstrated in the cone specimens.

TABLE VIII. FINAL DIAGNOSIS

Dysplasia	19
Carcinoma in situ	163
Invasive carcinoma	18
Total	200

Comment

The histologic study of the material presented in this report would indicate that the great majority of lesions described as dysplasia, carcinoma in

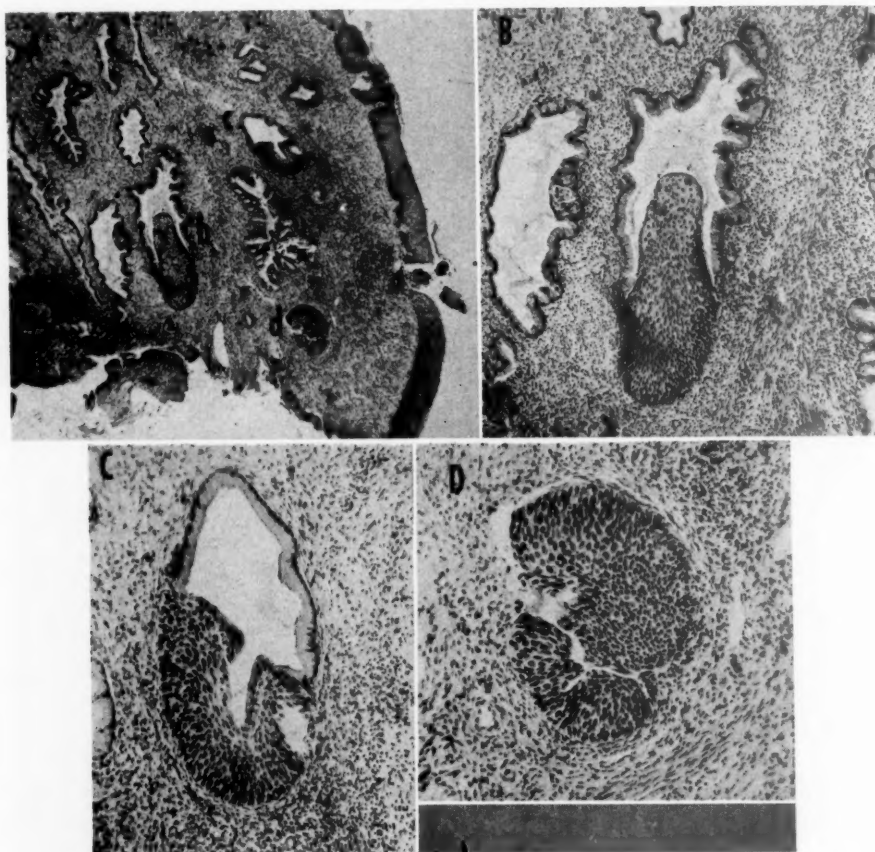


Fig. 4.—A, Three gland spaces deep within the stroma showing dysplasia of the basal cells. ($\times 38$; reduced $\frac{1}{2}$.) B, Higher magnification of gland b in A. ($\times 95$; reduced $\frac{1}{2}$.) C, Higher magnification of gland c in A. ($\times 175$; reduced $\frac{1}{2}$.) D, Higher magnification of gland d in A. ($\times 195$; reduced $\frac{1}{2}$.)

situ, and invasive squamous cell carcinoma of the cervix uteri arise from the basal cells of the epithelium of the endocervix and endocervical glands, as shown in Tables II through VI. The squamous epithelium of the portio vaginalis would not seem to be a frequent primary site of origin of these dysplastic and neoplastic changes. The potential of the reserve cells to undergo hyperplasia and to differentiate to squamous epithelium was probably first illustrated by Carmichael and Jeaffreson.^{1,2} This concept is in contrast to

the theory suggested by Meyer,⁹ who maintained that very early squamous cell carcinomas always arise from squamous epithelium. The potential of the basal cells to undergo hyperplasia and to differentiate to squamous epithelium is illustrated in the series of photomicrographs shown in Fig. 3. The dysplastic potential of these cells is illustrated in Fig. 4. Deep within the stroma of the cervix are illustrated three glands in which these changes are shown.

Fig. 5 illustrates carcinoma in situ arising from the basal cells of endocervical glands. These series of illustrations, showing various changes in the

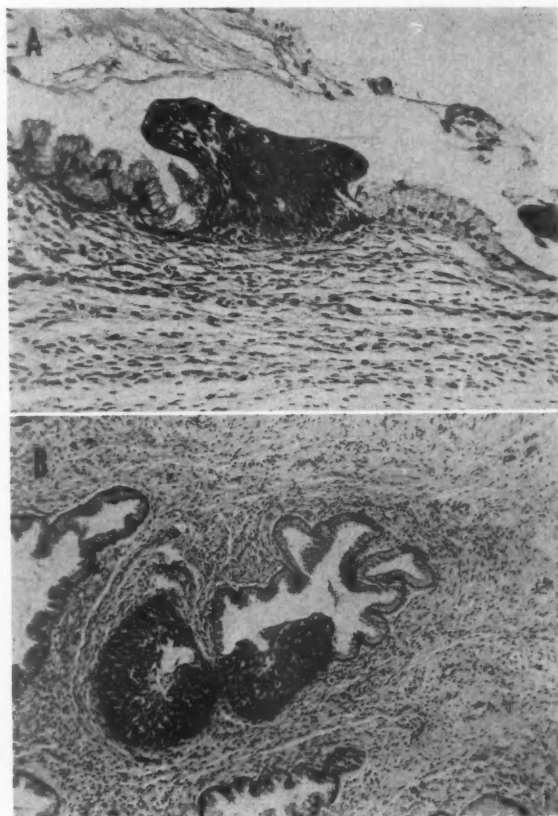


Fig. 5.—A, Carcinoma in situ involving gland space and probably by transformation of reserve cells. ($\times 240$; reduced $\frac{1}{2}$.) B, Section from cone shown in A. One gland space is filled with carcinoma in situ and another shows limited involvement of glandular epithelium. ($\times 110$; reduced $\frac{1}{2}$.)

basal cells, should not be construed as representing progressive stages in the development of carcinoma in situ from the basal cells of the endocervical epithelium. They are presented to demonstrate the varied potential of these cells.

Multicentric foci of neoplastic changes are shown in Fig. 6. The illustrations shown in these three sections were made from a radical hysterectomy specimen of a clinical Stage I lesion of the cervix, and this specimen is not included in the study. The anterior vaginal cuff is shown in the upper right corner of Figure 6, A. Cervical glands are seen far out on the portio

with squamous epithelium on each side of glands involved by in situ carcinoma. Early invasive carcinoma with glandular configuration is shown in the left lower corner of this illustration.

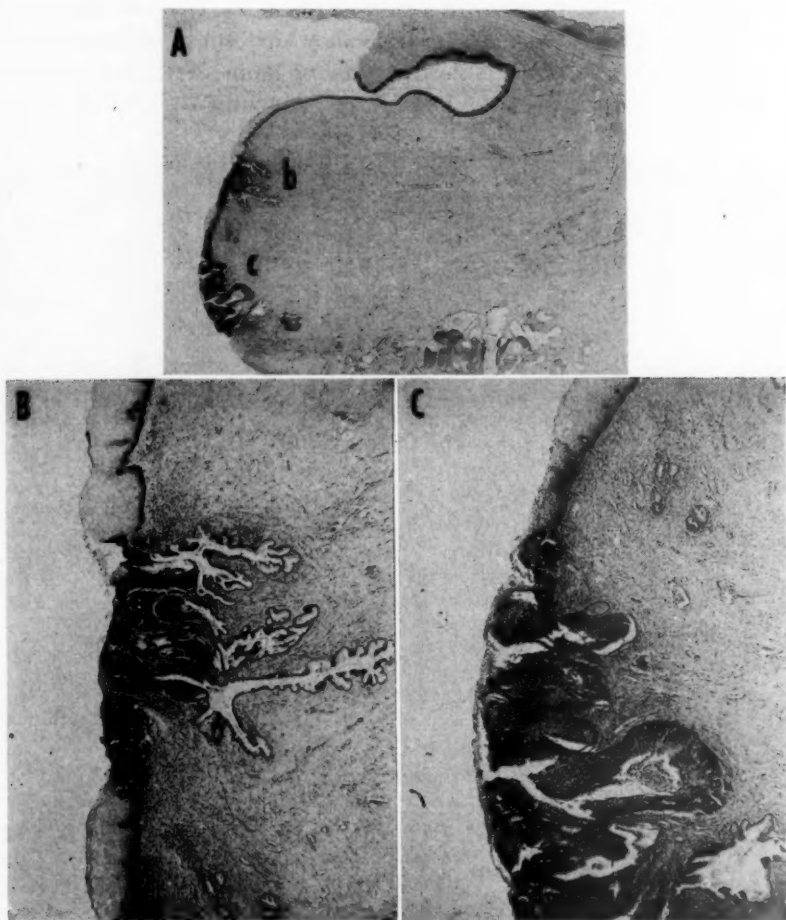


Fig. 6.—A, Panoramic view of anterior lip of cervix and vaginal cuff of section from a radical hysterectomy specimen showing carcinoma in situ in area *b*, and invasive cancer in area *c*. ($\times 8$; reduced $\frac{1}{4}$.) B, Higher magnification of area *b* in A, showing carcinoma in situ involving glandular epithelium. Squamous epithelium clearly seen on either side of the lesion. Lesion is far out on portio of cervix. ($\times 30$; reduced $\frac{1}{4}$.) C, Higher magnification of area showing early invasive carcinoma in area *c* of A. Other sections from this specimen showed extensive stromal invasion. ($\times 32$; reduced $\frac{1}{4}$.)

Summary

The histologic findings in 200 cold-knife cone specimens of the uterine cervix have been presented. The variations in the anatomic location of the transformation zone have been noted, and these variations are shown in a series of schematic illustrations. In 28 specimens it was not possible to identify the squamocolumnar junction for reasons specified. The dysplastic and neoplastic lesions were found in the region of the transformation zone in

all except 11 specimens of 172 in which it was possible to plot this zone. The epithelium of the endocervix or the endocervical glands was involved in 193 of the 200 specimens (96 per cent). From these studies it would appear that the dysplastic and neoplastic processes, in all probability, began in the basal cells lying beneath the columnar epithelium of the endocervical canal or the endocervical glands. This study would suggest that the squamous epithelium of the portio vaginalis is an infrequent site of origin of these changes.

These findings are of significant clinical importance in the early detection of neoplastic disease of the uterine cervix. Because of the variation in the anatomic location of the transformation zone, it would appear that exfoliative cytologic studies are the most satisfactory methods of detecting significant cellular changes in this zone. The high percentage of specimens in which it appears that the reserve cells are involved in these dysplastic and neoplastic changes would indicate that atypical exfoliative cytologic changes are best investigated by a tissue specimen which not only includes the transformation zone but also a significant portion of the endocervix for histologic study of the epithelium of the endocervix and endocervical glands.

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Discussion

DR. KARL H. MARTZLOFF, Portland, Ore.—Dr. Thornton's presentation offers an interesting continuation of the search to determine the site of origin of cervical cancer and to attribute some special significance to the area about the external cervical os where columnar mucosa and multilayered epithelium meet.

This is a far more precise attempt at site fixation than that used by many earlier writers, when the term "portio cancer" was applied to tumors arising on the vaginal aspect of the cervix, and "cervix carcinoma" was used for those arising within the cervical canal. It is interesting that this terminology has been used for years by Ruge, by Veit, and by von Franqué (1930) through successive editions of the *Veit-Stoeckel Handbuch*. During this interval (1900) Cullen's monograph appeared, followed in 1912 by that of Schottlaender and Kermauner. While the latter used this terminology, probably because it was so firmly established in the German literature, they did this with reserve and without emphasis. In fact, they favored dropping the segregation, because their own observations, in so far as portio cancer was concerned, did little to support it, and it is possible that they also derived encouragement from Cullen's complete avoidance of this apparently stereotyped classification.

As a further effort to localize the site of origin of cervical cancer we find that Schottlaender and Kermauner and Rosthorn, whom they quoted, observed, where this could

be determined, a small predilection for the tumor to involve the posterior wall. Because of the observation of patients in earlier stages of the disease than those studied by Cullen and Schottlaender, efforts have been made to determine with microscopic precision the site of origin of these cancers. The careful studies by Schiller (1927), von Franqué (1930), and others of suspicious as well as of unsuspected cervixes reflected this type of effort and permitted the recognition of cancer in its early preclinical phases. As a result, von Franqué and others have now demonstrated clearly that there is such a process as primary cancer of the portio and that the most common form of cervical cancer occurs in the environs of the external os in the *general* region of the epithelial transition zone where columnar and stratified epithelium form a shifting, unstable junction.

On the basis of deductions derived from more recent topographic studies it has become almost a "cliché" that most cervical cancers arise at the squamocolumnar epithelial junction and that tissue for biopsy removed from four quadrants about the external os would disclose most of these. This precise localization is based on the frequency with which intraepithelial changes (cancer *in situ*) were reported by some workers, particularly Foote and Stewart (1948), to occur in the vicinity of the external cervical os. The foregoing concepts, of course, assume that the external os and the squamocolumnar junction are topologically similar and that the "four quadrant punch biopsy" affords *ipso facto* an easy "diagnostic package."

Happily, Dr. Thornton has reconfirmed a number of basic observations, viz., that the external cervical os represents a macroscopic anatomical zone without precise histologic delineation; that normally variations occur in the anatomic location of the stratocolumnar junction; that the latter does not necessarily define the site of the external os; and that cancerous or precancerous changes may occur on any surface of the cervix or vagina.

Our own experience as to the general topographical distribution of the atypias, the intraepithelial changes of cancer *in situ*, and the small clinically covert cancers corresponds in general with Dr. Thornton's and with those of Fennell and particularly Przybora, whom he quotes. Statistical variations that occur on this point have little significance, I believe, since the bulk of small cancers or intraepithelial cancerlike alterations occur in the cervical canal and in the general region of the external os.

Time does not permit discussion of the role played by the so-called subcolumnar reserve cells, which Dr. Thornton considers the prime source of cells involved in these neoplastic changes or the factors involved in the development of squamous cell metaplasia.

However, there is a consideration in Dr. Thornton's presentation which is not clear to me. That is his reference to the precise relationship of "the anatomic location of the stratocolumnar junction" to the neoplastic changes under discussion. This is a tangible topographic and morphologic consideration to which I will confine the remainder of this discussion, since it does not involve the varying views and theories concerning the derivative cytology or genesis of cancer cells. Is this "junction" the starting point for most of these changes as is so commonly implied in general discussions? Personally, I have never observed this precise localization and I am now confining my remarks to the endocervical epidermoid neoplasias and to the point where multilayered cervical and columnar epithelium meet.

Merely for clarification, it may be helpful to mention a few considerations about which there is general agreement: (1) cancer of the cervix may occur in any part of the organ; (2) epidermoid cancer is the most common variety, although it frequently arises in the cervical canal where columnar epithelium normally occurs; (3) multilayering (squamous cell metaplasia) of the columnar epithelium of the canal (surface and glandular) is common; (4) it may be limited, diffuse, or multicentric; (5) the latter also may be said for cancer *in situ*.

On purely morphologic grounds, one cannot escape the observation that one of the important findings in the localization of intraepithelial carcinoid changes involving the canal is not the squamocolumnar junction but is the fact that these changes occur in association with a multilayered epithelium of varying extent and distribution. One

may commonly observe discrete plaques of squamous cell metaplasia in juxtaposition to columnar epithelium in the average adult cervix so that a single specimen may have many stratocolumnar junctions not necessarily related to the zone about the external os. Cancer in situ may occur in the midst of such metaplastic squamous cell plaques with unaltered stratified epithelium surrounding it. The same picture may be observed in the cervical glands without any direct connection with these surface changes. To me the outstanding observation in a study of such material is that the cancerous or noninvasive cancerlike focus regularly involves multilayered epithelium, not the columnar and ordinarily not the stratocolumnar junctions. It therefore seems apparent that, since the intraepithelial changes of cancer in situ as well as cancer may arise in any portion of the cervical canal, the consideration of prime morphologic importance concerns the associated squamous cell metaplasia which, so commonly, replaces columnar epithelium. Evidently, as compared to unaltered columnar epithelium, this is the epithelial change that is particularly responsive to abnormal growth stimulation.

In conclusion I confess my failure to recognize and understand the peculiar role which the stratocolumnar junction is supposed to possess. It is interesting to note that Dr. Thornton in his study also draws no conclusions concerning this.

DR. BAYARD CARTER, Durham, N. C.—Drs. Thornton, Fox, and Smith state that their study was concerned with the relationship of the squamocolumnar junction, or transformation zone, and the types of epithelia of the uterine cervix in the development of carcinoma in situ and of invasive cancer.

Their historical review is adequate and reminds me of the work done on the epithelia of the cervix by Rubin; Martzloff; Foote and Stewart; Meyer; Carmichael and Jeaffreson; and Howard, Erickson, and Stoddard. Meyer's study showed that the zone of mixed epithelium did not appear to be static. The morphologic potential of the small, undifferentiated basal cells, or reserve cells, which lie beneath the mucous epithelium of the endocervix and of the epithelium of the endocervical glands, is most important. These reserve cells may undergo hyperplasia and may differentiate to squamous epithelium. We are familiar with the potential of these basal, or reserve, cells because the tissue for the studies of Howard, Erickson, and Stoddard came from patients in our obstetric and gynecologic clinics upon whom we were doing the clinical studies of carcinoma in situ and of early invasive cancer.

The authors' study was a histologic study of 200 cold-knife cone specimens of the cervix. In 172 specimens the squamocolumnar junction could be identified and the cases were classified into the four groups which are shown in the illustrations.

It should be noted that they state: "It was not possible to plot the squamocolumnar junction in 28 specimens because of: (a) previous multiple punch biopsies, (b) previous cauterization of the cervix prior to adequate investigation, (c) faulty technique in obtaining the cone specimen, or (d) poor technique in preparing the sections in the pathology laboratory."

This statement forcibly stresses the necessity of proper precautions in the cone biopsy technique. Punch or wedge biopsies, cauterization of the cervix, and sounding and curettage of the cervix and of the uterus through the dilated endocervix should be prohibited prior to taking the cold-knife specimen.

Of the 172 specimens which were acceptable for study, the portio squamous epithelium was involved in but 6. Of the 28 cone biopsies discarded from the study because of the reasons given previously, the portio squamous epithelium was involved in one specimen.

Of 172 specimens, the change was found in the region of the transformation zone in 161. In only 11 specimens was the lesion at a great distance from the zone of mixed epithelia.

DR. RICHARD W. TE LINDE, Baltimore, Md.—The author has very properly emphasized the importance of cytologic study plus conization in determining the exact

nature and origin of carcinoma in situ. However, there is another step to this study which I have had in mind for a long time and I give it as a suggestion. You heard Dr. Fluhmann's presentation yesterday. That same type of study by plastic plate modeling should be applied here. If you see carcinoma in situ down in the depths of a gland, you see it in just one section. There should be a study by reconstruction to determine the relationship of that to the cervical epithelium. That would be a second chapter and there would be great value in such a study.

From a practical standpoint, I do not think we should concentrate so much of our attention on the cervical canal that we forget about the portio, although strictly on the surface carcinoma in situ may be widespread. In 275 cases of carcinoma in situ that we have treated with hysterectomy, we have had about 7 recurrences in the vaginal vault. Our treatment has been criticized as being too radical, and I am sure that some cases of carcinoma in situ can be cured by conization, but in a few instances our treatment has not been radical enough.

Although we have not in the distant past used Schiller's test as often as we should have, we now do it routinely, and I would recommend routine Schiller tests to guide you as to the amount of cuff that should be removed and thus prevent some recurrences in the future. In the recurrences in our series it seems obvious that we did not remove all the carcinoma in situ at the primary operation.

DR. ROGER B. SCOTT, Cleveland, Ohio.—I would like to add support to Dr. Thornton's demonstration inasmuch as Dr. Reagan, a competent cytologist and pathologist, and I have felt very strongly that we have been able to demonstrate very much what he has shown, viz., that the major epithelial alteration is found in the canal in most instances. We ran a large series of cases cytologically to determine which was the best comprehensive cellular sample obtained from the cervix. We studied vaginal pool aspirations as one unit, scrapings from the cervix as a second unit, and scrapings from the cervix and aspirations from the cervical canal as a third. The best comprehensive sample was the last—the combination of scrapings from the cervix and the aspirations of the cervical canal. To further prove that the major epithelial alteration is within the canal, we had 100 specimens of carcinoma in situ that we were able to study fairly completely because of the fact that the entire uterus or a wide and deep, sharp cone was available. We found in 90 of these 100 specimens that the major epithelial alteration was within the canal. This, I think, is of considerable importance. Unless we obtain a comprehensive cytologic specimen we cannot blame the cytologist for misinterpreting or underestimating the lesions actually present. We have to obtain the proper sample.

DR. LOUIS M. HELLMAN, Brooklyn, N. Y.—I, too, agree with what has been said, but I have some questions which have been on my mind for some time.

One can make up an extremely attractive story in regard to the reserve cell and its relationship to anaplasia, carcinoma in situ, and even carcinoma. This cell is stimulated in the cervix of the female fetus in utero by the maternal estrogen. Many of the changes which were shown by Dr. Thornton can also be found in the stillborn female infant. Furthermore, one can stimulate these changes in the reserve cell in postmenopausal women by administering large doses of estrogen for a short period of time. In these latter circumstances and in the ones mentioned earlier one finds hyperplasia of the glands and of the reserve cell with squamous and atypical changes. That these can be found under proper stimulation at both extremes of life is very interesting. One would like to hypothesize that in most instances the reserve cell, when stimulated, responds with a normal cell but where there is an inherited, inborn error the reserve cell exhibits an atypical response to estrogen and then if one adds an X factor one may get carcinoma of the cervix. This is perhaps what both Dr. Scott and Dr. Thornton have implied.

There are, however, some questions which can be raised against such an attractive hypothesis. In the first place the reserve cell is capable of several responses. It can form glands or squamous cells and yet in only one type of response of the reserve cell does

carcinoma appear, i.e., in the squamous cell response. Why is this so? Second, if the reserve cell is responsible for most carcinoma in the endocervix, it then must be admitted that carcinoma of the ectocervix comes from a different cell. These two carcinomas then should act differently in their development and perhaps in their prognosis but there is no evidence that this is so.

While I find the reserve cell theory most attractive, I would like to leave you with the idea that it may not be the answer to carcinoma and that a multicentric origin of cervical cancer, unrelated to this undifferentiated cell, may be a more correct hypothesis.

DR. THORNTON (Closing).—In answer to Dr. Martzloff's question about the significance of the squamocolumnar junction, all we wished to imply was that the anatomic or clinical location may vary with the histologic location. However, in spite of the variations, the majority of the lesions observed by us occurred in and about this zone. I believe we are discussing the same thing in regard to metaplasia and dysplasia of the endocervical and glandular epithelium.

Dr. Te Linde has offered some valuable suggestions which will be helpful in future studies of this problem. In regard to vault recurrence, one should not forget that when the uterus is removed for carcinoma in situ one removes a focus but may not have altered the stimuli responsible for these changes.

We are in agreement with Dr. Scott in that a spread which does not contain endocervical cells may not reveal to the cytologist significant changes which are present.

Dr. Hellman has pointed out the varied potential of the reserve cells as observed in his investigations. He has also presented questions which cannot be answered at this time.

RADICAL VULVECTOMY WITH BILATERAL INGUINAL, FEMORAL, AND ILIAC NODE RESECTION*

HOWARD ULFELDER, M.D., BOSTON, MASS.

(From the Vincent Memorial Hospital (Gynecological Service of the Massachusetts General Hospital) and the Pondville Hospital, Department of Public Health, Commonwealth of Massachusetts, at Walpole)

TO CURE cancer of the vulva, the surgeon must excise widely the entire vulva and the fat and lymph apparatus in both inguinofemoral fossae. Many men^{4, 12, 20, 22} believe that in addition the deep pelvic lymph nodes identified with the external and internal iliac vessels on both sides should always be removed; everyone would agree that this step is indicated when metastasis is already present in the superficial nodes. Most authors^{2, 9, 18, 23} have also taken pains to point out the inadequacy of lesser operations particularly unilateral vulvectomy or wide local excision.

The history of surgery for cancer of the vulva illustrates acceptance of these principles and their application with increasing courage as a variety of factors have evolved to make operations more safe in the elderly patient. Basset's¹ concept of the attack on the inguinal nodes, though inadequate in extent by present standards, was nonetheless a significant forward step at the time it was described because it advocated removal of tissues before gross disease could be felt in them but in anticipation of a high percentage of chances that cancer would next appear there. Both Kehrer⁶ and Stoeckel¹⁵ in Germany performed a wide resection of vulva and nodes in selected cases and specifically discussed the inaccuracies of clinical appraisal of the lymph nodes for metastasis.

The kind of factual evidence on which is founded our present idea of the extent of treatment necessary in these cases was first collected and presented by Taussig.¹⁶ He recommended and regularly practiced an adequate operation but found the morbidity with Basset's long bilateral inguinal incision too great for complacency. To reduce this, he advocated operation in stages,¹⁸ first doing the radical groin dissection on the side of the tumor, then a similar dissection in the other groin, and finally a wide vulvectomy. As he accumulated experience he stopped cutting Poupart's ligaments for exposure and made his inguinal incisions in the axis of the vessels rather than in the crease of the groin. He noted no increased danger of recurrence from leaving the skin bridge between the vulva and the groins.

Data to confirm the low cure rate following limited operations were also found by Way²³ when he collated the cases at Newcastle Infirmary. As a

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result, in 1942²² he first performed in one stage an operation deliberately designed to be adequate for the disease and which included a generous resection of the skin over the mons veneris and Scarpa's triangles. With this plan, his cure rate was excellent²⁴ but, of 75 patients so treated, 11 died in the hospital²⁶ and the average stay was many days. This mortality and morbidity Way ascribes to the large wounds which could not be closed per primum. A more recent report²⁵ describes his present procedure, an extension of the incision in the thigh which permits him to swing simple flaps to cover most of the open wound surface.

In this country, McKelvey clearly recognized the importance of Taussig's contributions and in 1938 inaugurated a schedule of management in his own clinic specially designed⁹ to permit wide and adequate dissection with a low risk. The important features of this program were the use of heavy sedation supplemented by local anesthesia, and one stage excision in continuity of the vulva and the contents of both superficial groins up to the peritoneum in the femoral and inguinal canals. There were 5 hospital deaths in 55 cases¹⁰ but none was the result of the extent of operation or its subsequent course.

Both McKelvey and Way discuss the immediate and remote problems created for the patient by this extensive operation. Necrosis of skin along the edges of the wounds is commonplace and as long as raw surfaces are exposed there is loss of serum sufficient in many cases to require replacement. The risk of invasive sepsis is always present. Free grafts of skin are used as soon as they are likely to take but under the best of circumstances recovery is slow. Both authors mention subsequent lymphedema of the legs as a frequent late complication; this is equally true of groin dissection for other diseases²⁰ or done in stages.²⁶

Strongly influenced by Taussig's reports, the treatment of cancer of the vulva in Meigs's clinics was planned in all cases to be bilateral and adequate.¹² Performance of the operation in stages kept the mortality low and a review of end results⁵ showed the same per cent of cures both with and without metastases as reported by Taussig, Collins, Way, and others. Dissatisfaction grew from 2 features. First, it was obvious that the length of hospitalization, the disability, and the expense involved for each patient were greater than for almost any other acute illness. Second and more important was the observation that a number of patients with potentially curable lesions never completed the full program because of long delays introduced by complications or for other less obvious causes. These problems were debated for several years and alternative incisions and sequences of staging were tried, aimed chiefly toward the goal of primary healing and consequent shortening of the over-all time under treatment. No real improvement was noted until November, 1949, when a patient presented with a lesion involving vulva, clitoris, and urethra, and I chose to do bilateral superficial groin dissections and simultaneous vulvectomy in continuity through a crescentic inguinal incision and vertical vulvar wound, both of which could be closed. Exposure of Scarpa's triangles was excellent (Fig. 1) and healing uncomplicated. One right femoral node showed metastasis and the patient returned in 2 months for bilateral iliac lymphadenectomy. The same crescent incision was used, again with good exposure and prompt healing. It was apparent that operation of adequate extent was practical by this approach and the majority of cases seen since that time have been managed in this way. The technical steps

of operation at present are almost identical with the procedure reported independently by Twombly²¹ and myself.¹⁴ It differs distinctively from Way's operation only in the lesser excision of skin, but we have not been tempted to make a modification in this feature because the effort has been to keep morbidity and mortality at a minimum, and our experience with staged operations has shown that only the skin usually removed by the radical vulvectomy need be sacrificed for cure of the disease.

Material and Methods

Between November, 1949, and September, 1958, 53 women with malignant tumors of the vulva have had radical operations through the crescent inguinal incision at the Pondville Hospital in Walpole, Massachusetts, and the Vincent Memorial Hospital (Gynecologic Service of the Massachusetts General Hospital) in Boston, Massachusetts. This presentation is an analysis of the records of these cases with the focus of attention deliberately on those details which will help

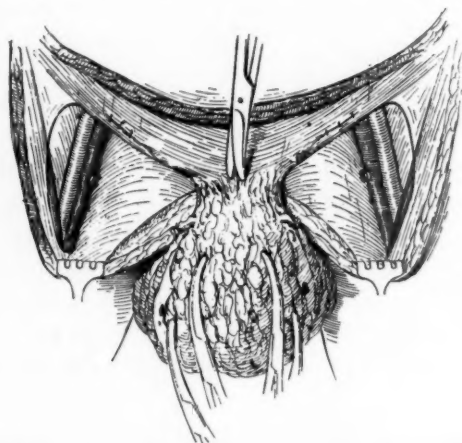


Fig. 1.—Crescent incision after bilateral inguofemoral node dissection. (From Parsons and Ulfelder: *An Atlas of Pelvic Operations*, Philadelphia, 1953, W. B. Saunders Company.)

us to understand the effect of this operation on the women who have accepted it. In other words—What price is being demanded of our patients when we offer to try to cure cancer of the vulva by this procedure?

This group of 53 patients is composed of individuals with the background history one has come to expect in association with cancer of the vulva: 38 of the women were over 60 years of age and 20 were over 70. There were 2 hospital deaths; one patient, 68 years of age, died suddenly of acute pulmonary embolism 5 days after the operation and another, 69 years of age, died of carcinomatosis 3 months after operation. In 23 patients the radical vulvectomy was combined with bilateral resection of the superficial nodes only. The average operating time in this group was 207 minutes, and the average blood replacement was 2 units per patient on the day of operation. In 30 cases where both the superficial and the deep nodes were resected at the time of primary operation, the average operating time was 235 minutes and the average blood replacement was $2\frac{1}{3}$ units. The usual indication for failure to carry out deep node dissection was increased operative hazard estimated either before or during the operation. However, the figures demonstrate so little difference between the two groups that we now feel the need for selection between these 2 operations is more apparent than real. It is at present customary to carry out the full procedure in almost every case.

For proper indication the deep nodes can be removed at a subsequent operation and this was done in 4 of the 23 patients listed above, when the pathologist reported unsuspected lymph node metastasis in the superficial nodes examined. Three of these 4 patients are living and well 6½, 7¼, and 9½ years later; the fourth patient died 18 months after operation of a cerebral hemorrhage but was apparently free of disease.

Postoperative Morbidity

In most cases the skin at the conclusion of the operation was closed, often under considerable tension. Usually there was some degree of necrosis of the skin flaps at the point where the transverse and vertical components met. With necrosis there was cutting of sutures and then retraction of the flaps before they again became adherent. This left a triangular area to granulate overlying the symphysis. Secondary operations to hasten healing of this area were carried out in 27 patients. Usually this consisted of multiple Thiersch grafts but on occasion it was possible by undermining the flaps to bring them together with secondary closure. In the other cases healing was accomplished without additional operation but in only 9 individuals was necrosis and separation so trivial that one can consider that healing occurred essentially per primum. In 42 patients the record indicates the length of time required before healing was finally accomplished. It averaged 75 days but the median length of time was 60 days, suggesting that a few patients with markedly delayed healing have raised the figure for the average length of time. Separate analysis of the cases with and without deep node resection reveals no difference in healing.

It is quite apparent from review of the records that these patients are considerably less disabled over the long-term follow-up than women who have had staged operations with long vertical incisions over the femoral vessels. In the present group of patients there are 16 in whom there is no complaint and no swelling noticeable on examination. In 9 patients there is mild edema of one or both legs, not at all incapacitating and not sufficient to justify the use of elastic stockings except for cosmetic reasons. Moderate edema which is noticeable and which requires some form of chronic care is present in 8 patients and in 4 women there is severe swelling of one or both legs.

Urinary stress incontinence is a complaint of 5 patients, very slight in 2 and moderate in 1. In the fourth patient there is definite incontinence but the patient states that this is about the same as it was before the operation. In the fifth patient, after a long enough postoperative interval to be sure that the tumor was probably cured, operation for the relief of incontinence was carried out and she is now in her ninth follow-up year without evidence of recurrent carcinoma and with good control.

Postoperative femoral hernia has not been the problem we anticipated. It has developed and been successfully repaired in 2 cases but we have balanced the ledger by carrying out successful repair of bilateral pre-existing femoral hernias at the time of vulvectomy in a third patient.

Comment

The immediate and late morbidity and mortality of the operation under scrutiny here has been shown to be less than for any previously reported group of patients given adequate surgical treatment for cancer of the vulva. The most striking single figure to support this statement is a calculated average postoperative stay of 44 days in the patients who had the complete operation including resection of the deep nodes and an average hospital stay

of 33 days in the patients who had radical vulvectomy and bilateral superficial nodes only. The most impressive clinical feature of these patients as they are examined subsequently is the absence of extensive ugly scarring and deformity in the region of the vulva and groins and the infrequent complaints of incapacitating peripheral edema. This latter fact we attribute to the absence of skin necrosis, granulations, and dense scar in the region of the femoral vessels.

The occasional patient that heals without complication, however, constantly encourages us to make minor modifications in technique. During recent years the crescent incision has been more transverse than formerly, with

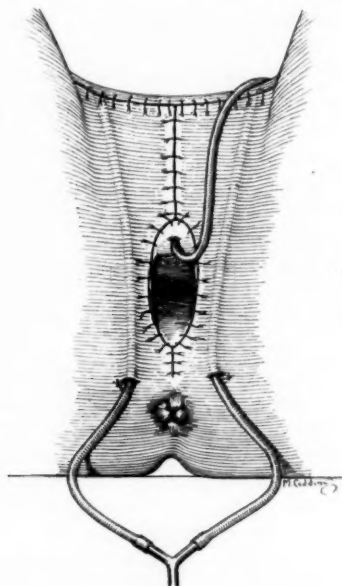


Fig. 2.—Appearance of wound after closure, with suction catheter drainage in place.

the ends below and lateral to the iliac spines and thus perhaps a little bit more in the line of the natural skin folds. This has allowed us to start the skin closure an inch or two medial to the corners of the upper flap and thus to convert a part of the original upper margin of the incision into lower margin during the closure and to compensate in some degree for the loss of skin at the site of vulvectomy. It has been noted also that closure of the vertical portion of the incision is accomplished with less tension if the patient's legs are lowered from the lithotomy position to the supine after the vulvectomy has been completed. Most recently the technique of drainage of the wound has been changed from soft rubber wicks to suction catheters.⁷ With these, large collections of fluid elevating the flaps no longer occur (Fig. 2).

Conclusion

The operation herein described is appropriate for treatment of cancer of the vulva in that it may be done in one sitting with little hazard even in

elderly and poor-risk women. The patients must expect to spend 4 to 6 weeks in the hospital and it may be another month before healing is complete. Unpleasant late deformities and disability are infrequent.

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Discussion

DR. CONRAD G. COLLINS, New Orleans, La.—We agree with the author that to cure invasive cancer of the vulva, removal of the entire vulva plus other procedures has to be utilized. Cancers proved to be noninvasive by multiple sections of the widely excised vulva do not require regional node dissections. Since 1946 we have so managed 13 such cases. Seven patients were operated upon 5 or more years ago. None have died of cancer and only one of the 13 developed a recurrence 1 mm. in diameter at the suture line. This recurrence was removed by wide excision. As regards invasive cancer we believe that unless there is distant metastasis to bone or lung the patient should be given the benefits of operation. This includes at least wide vulvar excision, bilateral resection of the superficial and deep inguinal, femoral, Cloquet's, external iliac, hypogastric, obturator, common iliac, and lower aortic and vena caval nodes. When contiguous structures such as the urethra, rectum, anus, levator ani muscles, or bladder are involved, the scope of vulvectomy should be extended to include removal of the involved structures even though anterior, posterior, or total exenteration is necessary. Of course, in the latter cases the extent of node dissection is the same as described previously.

Since 1946 all cases of vulvar cancer observed on our service have been cared for by the therapy prescribed by one of the 3 members of our vulvar team. Dr. Ulfelder has not informed us whether or not his series consists of consecutive cases, such as ours. In other words, has anyone been denied the benefits of operation because of extent of lesion, age, or associated medical conditions? To reiterate, irrespective of any other factor, on our service, operation is performed unless there are distant metastases to lung or bone. In our series of 56 patients, 49 had removal of vulva and nodes, 7 had exenteration. In the 49 patients having removal of vulva and nodes 15 additional procedures, such as removal of vagina, uterus, cervical stump, urethra, rectum, or levator ani muscles, were performed. In this series 18 patients were over 70 years of age and 30 were over 60 years of age. This is comparable in number to the author's series.

We do not believe that the type of incision has anything to do with mortality, morbidity, or cure. Fat persons do not heal as well as thin persons no matter where an incision may be placed on their bodies. Furthermore, in the aged, wound healing is not as good as in the young, and though the aim of every surgeon is to have his incisions heal by primary intention such cannot always be accomplished. In operation for carcinoma of the vulva, whether one is to obtain healing of the vulvar area by primary intention or not is of minor importance. Of major importance is to excise the vulva widely and deeply enough to remove the cancer with a good margin of normal tissue. As regards the method of removal of nodes, whether by longitudinal incision or crescentic incision, we do not believe this is important. We believe we can obtain better exposure of the area in which we choose to work by utilizing the longitudinal incisions. Primary wound healing in the vulvar area occurred in 70 per cent of our cases. In only 3 was skin grafting considered. Conversely, in node dissection incision, in only 16 cases was primary healing attained. Of late we have been utilizing catheter suction beneath the fat for 48 to 72 hours post-operative but have not had enough cases as yet to say whether or not this has materially reduced the longitudinal incision breakdown.

As regards staged procedures, whether one does the operation in one or two stages is of no import. The second stage can be performed a few days after the first. What one is attempting to do is to cure cancer. A two-stage operation does not materially increase the patient's hospital stay. Our average hospital stay is 32 days. Our average operating time for residents is 4½ hours and for staff 3½ hours. Therefore we do not see any advantage of the crescentic type of incision over the bilateral longitudinal incision. As regards late results, there were 28 patients who had no trouble with their legs. Of the remainder, 18 had edema in the immediate postoperative course. Ten had continuing edema from time to time, but only 3 of these had continuing difficulties, usually recurrent superficial phlebitis.

In conclusion we believe that invasive cancer should be treated by wide vulvectomy, superficial and deep pelvic node dissection, and removal of affected continuous structures. The type of incision for removal of nodes is of no import as we cannot see any advantage or disadvantage in utilizing the procedures of bilateral longitudinal incision with or without cutting or removal of Poupart's ligament as contrasted to the incision described by Dr. Ulfelder. Nor is there any difference in the early or late results.

DR. FRANK R. LOCK, Winston-Salem, N. C.—Although our experience with one-stage radical vulvectomy with inguinal, femoral, and iliac node dissection is somewhat limited, we are much more inclined to this approach than we were in the past. The two-stage procedure does not appreciably reduce the morbidity or mortality and the total hospitalization is increased.

A complication we experienced should be reported since it is common to each approach and represents an unexpected emergency in gynecological practice. We are all aware of the frequency with which separation of the wound occurs with these procedures, particularly in the area of groin dissection. Exposure of the femoral vessels may result so that they are exposed in the granulating wound for a considerable period of time. In a similar situation with radical dissection of the neck, rupture of the carotid vessels occurs with some frequency.

Approximately one month ago spontaneous rupture of the left femoral artery occurred on the twelfth postoperative day in a 78-year-old woman upon whom I had performed a radical vulvectomy and regional lymphadenectomy for squamous cell carcinoma of the vulva. The wounds in the groin had been treated postoperatively with continuous suction and hypothermia which was produced with ice packs. The wounds separated in spite of this and the major vessels were exposed for a distance of 5 to 6 cm. Wet dressings were applied. The wounds appeared clean.

A resident was in the patient's room when blood spurted from the wound. He applied his finger to the vessel until the patient could be transported to the operating room and transfusion of whole blood begun. The femoral artery had ruptured just below the profunda branch, so the circulation to the leg was satisfactory after ligation of the femoral vessel and secondary closure of the wound. Sympathetic nerve blocks were maintained for 72 hours. The patient survived and has satisfactory function of the leg without trophic changes.

Although we have not favored secondary closure of these wounds in the past when they broke down, I am convinced that the major vessels should be covered with adequate tissue by a secondary procedure if necessary after this experience.

DR. ERLE HENRIKSEN, Los Angeles, Calif.—I have debated with my conscience whether to be completely honest, or, by quoting statistics, be scientific. Honesty won out, and thus I base my comments on the careful study of the lymph nodes in 7 patients with carcinoma of the vulva who died of other causes. It is my impression that the non-malignant histologic character of the superficial nodes is of extreme importance. In our small series, the history of a long-standing ulcerative vulvar lesion is usually associated with marked changes in the node structure. In such cases, a "hit-and-run" reaction need be considered. The malignant cells strike the scarred node, their expected route of travel is obstructed, and it is possible for these cells to travel in an opposite direction. In 3 of the 7 cases, with abnormal, but nonmalignant changes in the nodes, positive nodes were found halfway down the inner thigh. Even the most radical approach would have missed these foci. In 2 cases in which the lesion actually involved the vaginal mucosa, positive perirectal nodes were present. Therefore, the loose and widely misused term "cure" should be replaced by a basically more descriptive term. It is most evident that in many of these cases, our present admittedly incomplete knowledge of the natural history of this disease clearly predicates not a cure but a temporary respite. To hold with the premise that operation can remove all of the nodes is certainly wishful thinking. Too often operation is carried beyond the realm of good judgment.

In my mind there is no argument between the single- and the two-stage approach. Admittedly, one cannot set rigid rules, but when possible, the one-stage operation is the one of choice. Cancer cells not only can be present in the major lymph node groups, but, to get there, they need travel via the channels, many of which have innumerable so-called anomalous nodes. To cut through these potentially involved channels may not only nullify any possible good arising from the operation, but may also enhance the danger of further dissemination.

DR. JOHN PARKS, Washington, D. C.—Since edema is the most important contributing factor to postoperative pain, there should be some way of taking care of this condition. About 8 years ago we started applying ice collars to episiotomy wounds to prevent edema and the procedure was so successful that we began to apply ice collars and ice bags immediately following vulvectomy. This has reduced tremendously the amount of pain and the degree of wound separation associated with vulvectomy.

DR. WILLIS BROWN, Little Rock, Ark.—I should like to read one very brief quotation from the program: "Carcinoma of the vulva has been shown to be a very curable lesion if *treated adequately*." I should like to suggest that carcinoma of the vulva has been shown to be a very curable lesion if *found early*. I am wondering if all the improved results

that we claim for surgery are not instead an improvement in our communities. As our citizens are educated to come to us earlier, better results will obtain and such heroic surgical efforts will not be necessary.

DR. JOHN L. McKELVEY, Minneapolis, Minn.—One must disagree with Dr. Brown. This is a curable lesion. Not only that but it is curable after a relatively long period of symptomatology in an area where it is easily recognized. With this in mind I would suggest to you something that has been of value to us. By the use of the rectospectroscope we have set up a clinical classification of vulvar carcinoma. Oddly enough, our Stage I lesion is one which approximates that described by Dr. Collins. It is a lesion 10 sq. cm. or less in area, which does not involve the urethra, vagina, or anus. In this stage it is, for practical purposes, completely curable by single-stage radical vulvectomy, stopping at the peritoneum of the inguinal and femoral canals. Stage IV means distant metastases and these are at present incurable. Stage II is a lesion larger than 10 sq. cm. without involvement of urethra, vagina, or anus, and of this group approximately two thirds will be alive at the end of 5 years. We can treat these Stage II lesions with some assurance of success by less than the very radical procedures that have been described.* Until a few years ago there were in our series no cures from these Stage III lesions. This meant one was dealing with an all-or-none problem. Various degrees of exenteration procedures are justified for Stage IV lesions and these have been done. This has produced some patients with reasonable expectation of cure. It has been of value to break the general rule of the desirability of a standardized procedure for the general group.

With the techniques presently available, it is expected that a primary healing of the inguinal and pubic wounds will be obtained. One should not limit the extent of the vulvar excision itself with the object of obtaining primary healing. The vulva will heal quite satisfactorily by secondary intention. A major primary consideration is the evidence of local vulvar tumor persistence or recurrence and this can be achieved only by very wide vulvectomy.

In all of these arguments about surgical techniques one must not lose sight of the basic fact that there is nothing as effective in producing cures as getting the lesions early. It is important to establish in the minds of the practitioners who first see the patients the conviction that vulvar complaints should be investigated and that they can be safely treated in even very elderly women by surgical means which are effective.

DR. ULFELDER (Closing).—I will not try to answer the individual discussers, but I would like to emphasize several facts which the discussers brought out and which have certainly helped to point up different features of my own collection of material which I did not have a chance to present to you today.

I do agree and I am heartened to hear that the important feature to all is that we must never feel that any patient with carcinoma of the vulva should be considered for what we feel is less than adequate treatment for the disease. There is some difference of opinion as to what adequate treatment involves. Dr. McKelvey feels that superficial node operation in this particular carcinoma would be justifiable when the lesion falls in one of his groups where less than completely radical procedure is necessary for cure. At one time we also flirted seriously with doing only superficial node dissection in all cases at the time of radical vulvectomy and then offering, at a second stage, bilateral deep resection if positive nodes were found. We found no difference in results whether we did or did not do the deep node dissection at the time of the primary procedure, and that is the basis for our program at the present time.

I am very grateful for the suggestions that will help in the management of our patients. We are using suction catheters for drainage under the flaps, using atmospheric pressure to keep the skin in close contact with the underlying tissues by having an airtight closure. It makes a good deal of difference in the end result. They have much more rapid healing and less loss of skin.

*Stage III is a lesion of any size, although it is usually large, which involves the vagina or anus as well as the urethra.

CELLULAR CHANGES IN VAGINAL AND BUCCAL SMEARS AFTER RADIATION: AN INDEX OF THE RADIOCURABILITY OF CARCINOMA OF THE CERVIX?*†

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IT IS probably safe to say that most clinicians who treat patients with carcinoma of the cervix are convinced that individuals with apparently similar tumors may or may not be cured by equal radiation therapy. This difference is ascribed to the somewhat intangible quality referred to as radiosensitivity.

There are many articles describing efforts to categorize and measure radiosensitivity in terms of various microscopic properties in tumor cells. This has recently been reviewed by Merrill¹ and need not be detailed at this time. Reference must be made, however, to the work of Ruth Graham, who was later joined by John Graham,² in developing a concept and a technique for predicting radiocurability of patients by examination of exfoliated vaginal cells. These workers noted that certain morphological changes could be observed in the normal vaginal cells following radiation. These changes were called radiation response (RR) and a correlation between a critical percentage of cells showing radiation response and radiocurability was noted.

The implication from this work is clear and important. If it would be possible to pretest the susceptibility of a patient and, therefore, presumably her tumor to radiation, it should be possible to ascertain beforehand the best therapeutic modality for that patient. Thus, radiation and operation would not be competitive modes and a preferred choice of treatment might be indicated through an objective diagnostic test.

The present investigation was begun in 1955 in an effort to repeat the findings of the Grahams. The first part of the results detailed below is devoted to a presentation of our results with their technique. Early in the program certain difficulties—practical and theoretical—became apparent.

It is to be recalled that the determination of radioresponse is made by examination of the normal vaginal cells after radiation. The differentiation of a malignant cell from a radiated normal cell is not always certain. The

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Grahams have been long aware of this problem and there is no doubt that accuracy improves with experience. Our own difficulties with this problem, however, were crystallized by the ease with which several experienced observers were confused by the slides of a patient without carcinoma who received a routine cervix radium application for cancer. We therefore have reservations about the ability of less-than-expert observers to always be sure on this point and have no doubt that it is a source of some error in our own counts.

The determination of radiation response in exfoliated vaginal cells is additionally open to the problem of cell distribution and selection with respect to the uncertainty of dosage received by a particular exfoliated cell. A radium application in the cervix subjects the normal cervical and vaginal cells to a range of several hundred to several thousand roentgens. As the final prognosis is dependent upon the per cent of cells exhibiting radiation response, the desirability of a standard irradiation dosage to the test cells is obvious.

In addition, vaginal smears are subject to ovarian hormonal influences which undergo profound changes during irradiation.

Finally, there is the intangible observer bias in applying the diagnostic criteria to particular cells. This, no doubt, can be overcome by persistent and prolonged training but it must be recognized as a practical difficulty in the widespread application of the method.

As might be suspected from these considerations, the Graham method has been the subject of some controversy in the literature. Unfortunately, up to the present there are few series with completely satisfying information which either confirms or fails to confirm the Graham technique.

The data of a number of workers have been unable to confirm the method but it must be said that many of these contain only preliminary information.³⁻⁹ On the other hand, several workers have confirmatory data, but here again the information is preliminary and incomplete except for Kjellgren who has beautiful confirmative data.¹⁰⁻¹³ Smith and associates,¹⁴ using a modification of the technique, have also presented preliminary confirmatory data.

Regardless of the cause, it seems clear that there is difficulty in easily transferring the method to other laboratories. Nevertheless, the concept of radiosensitivity testing is of such therapeutic importance that we have continued to investigate the problem, using a somewhat different technique.

Except for the Grahams, all attempts to define radiosensitivity, such as those of Martzloff,¹⁵ Cherry and Glucksmann,¹⁶ Gusberg,¹⁷ and others, have used the tumor as the test object. One of the Grahams' important contributions, however, has been the concept that the response of normal cells to radiation may be used as a measure of tumor response, at least with respect to carcinoma of the cervix.

In order to overcome the problem of cell identification, to aid in minimizing the endocrine influence, and to assure a standard radiation dosage, the test site was moved away from the tumor and a standardized dosage of roentgen rays was administered to a uniform area of the buccal mucosa. Following this, the changes in the cells gently scraped from the test area at

intervals following the test irradiation were observed. The cell changes have been tabulated according to a number of criteria and the results of such analyses compared with the radiocurability of the patient. The number of

Fig. 1.

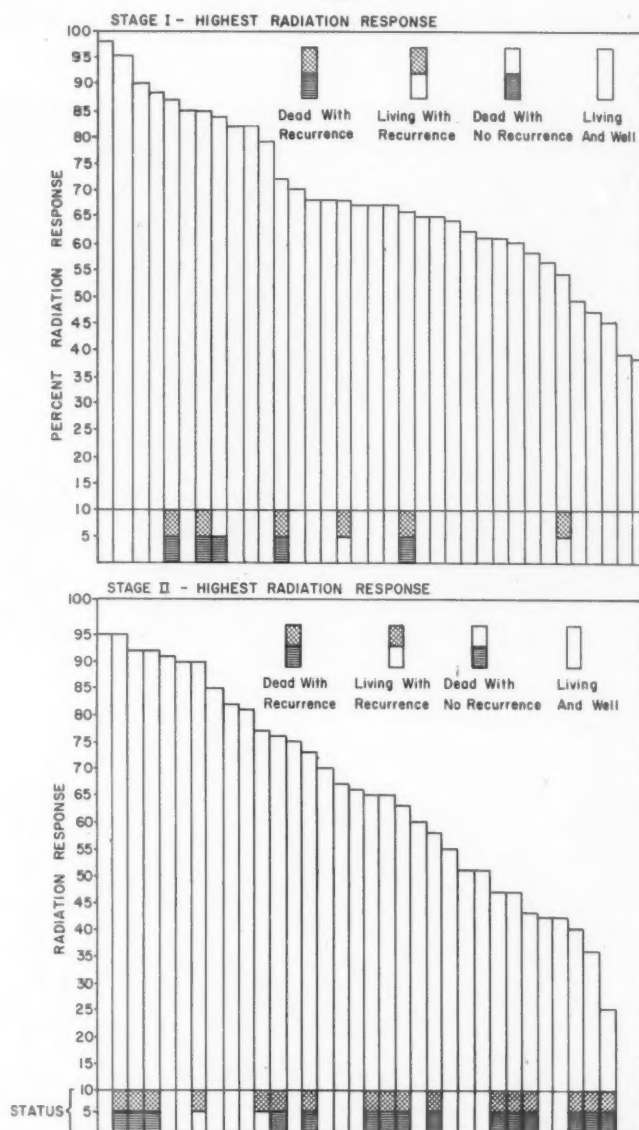


Fig. 2.

Fig. 1.—Vaginal smear. Highest radiation response and 2 year end result in 35 patients with Stage I carcinoma of cervix.

Fig. 2.—Vaginal smear. Highest radiation response and 2 year end result in 33 patients with Stage II carcinoma of cervix.

patients tested, the necessary retrospective nature of the experiment, and the length of the follow-up period make it necessary to emphasize that the conclusions must of necessity be regarded as tentative at the present time.

Estimation of Radiocurability by 4 Morphological Characteristics of Exfoliated Vaginal Cells After Radium

This constitutes a report of 68 patients with squamous cell carcinoma of the cervix treated with radiation and followed for at least 2 years. During radiation, serial vaginal smears were obtained 3 times a week and examined

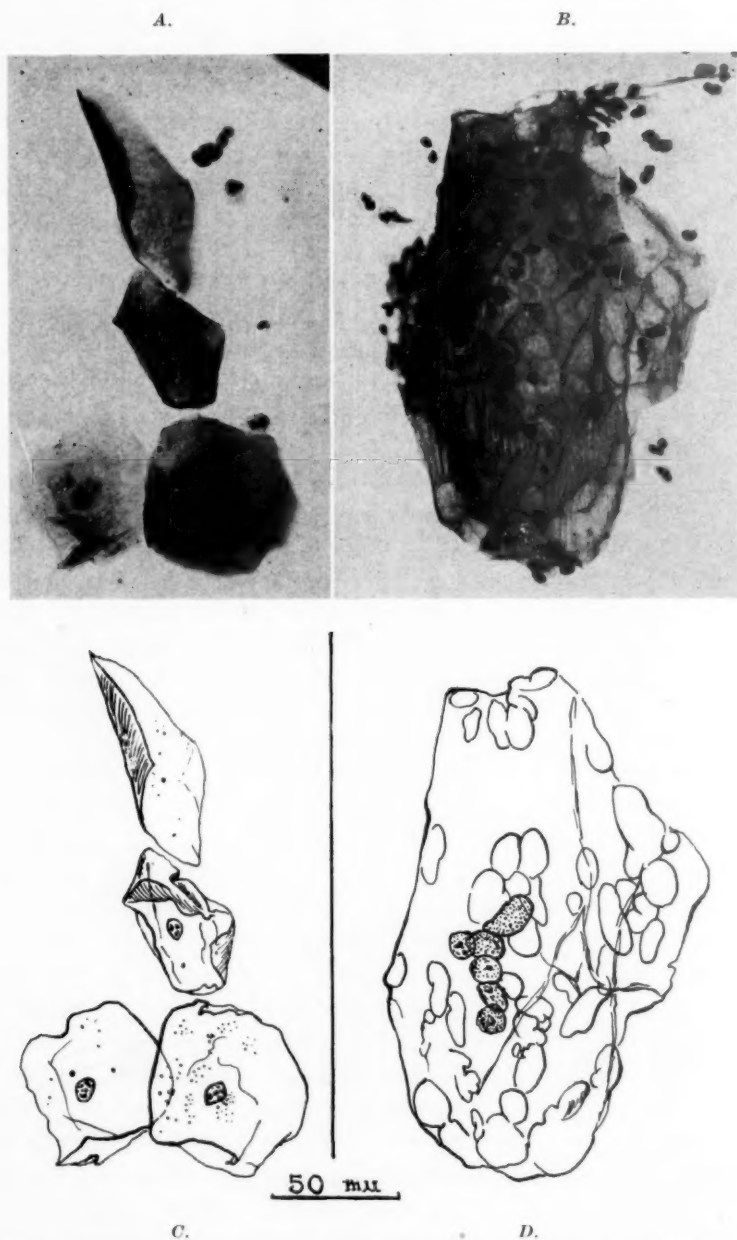


Fig. 3.—A, Control buccal smear. ($\times 500$; reduced $\frac{1}{2}$.) B, A 12 day cell of a buccal smear from a highly responsive patient. Great cell enlargement, cytoplasmic vacuolization, and multinucleation are well shown. Nuclear chromatin clumping is not clearly seen. ($\times 500$; reduced $\frac{1}{2}$.) C and D, Pen and ink sketch emphasizing cellular features.

according to the method outlined by Graham.² The significance of the 2 year follow-up as compared with the conventional 5 year period will be discussed below.

There were 35 patients with Stage I carcinoma of the cervix. Of these, 12 had good irradiation response and 9 (75 per cent) were living and well at the end of 2 years. There were 23 patients with poor response and 19 (82 per

Fig. 4.

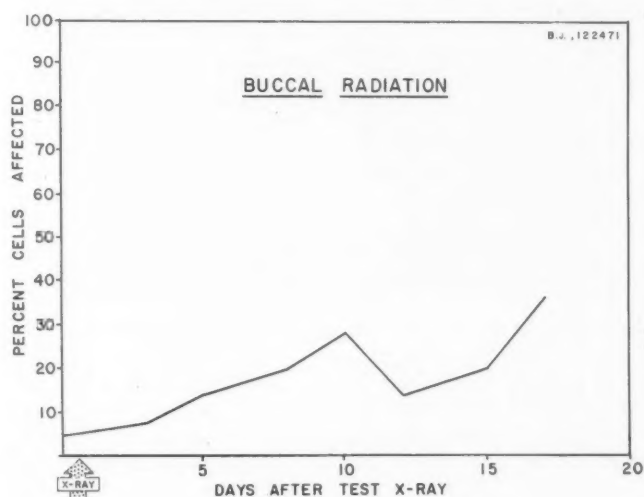
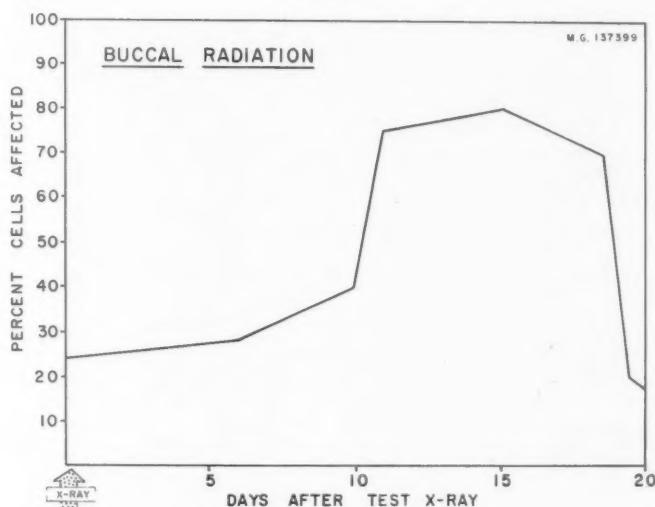


Fig. 5.

Fig. 4.—Curve showing per cent cells affected by radiation according to days after test dose. A good response curve.

Fig. 5.—Curve showing per cent cells affected by radiation according to days after test dose. A poor response curve.

cent) were living and well at the end of a similar period (Fig. 1). On the basis of this experience, it is necessary to conclude that for Stage I carcinoma of the cervix there was no difference between the good and the poor radiation response groups.

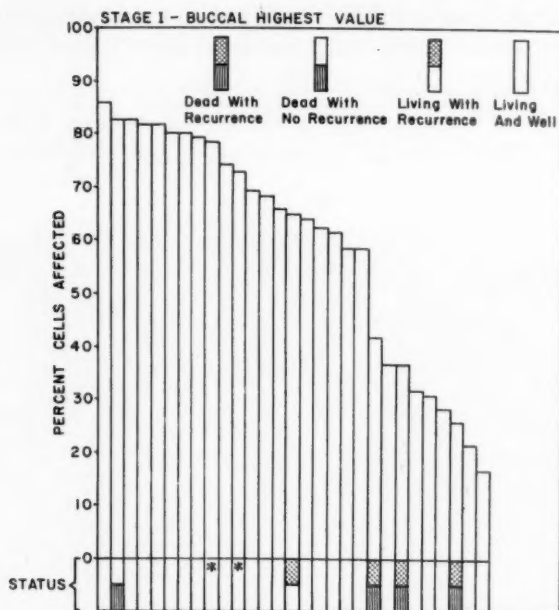


Fig. 6.—Buccal smear. Highest radiation response and 2 year end result in 29 patients with Stage I carcinoma of cervix. The 2 asterisks identify patients who had recurrences 7 and 13 years, respectively, after treatment.

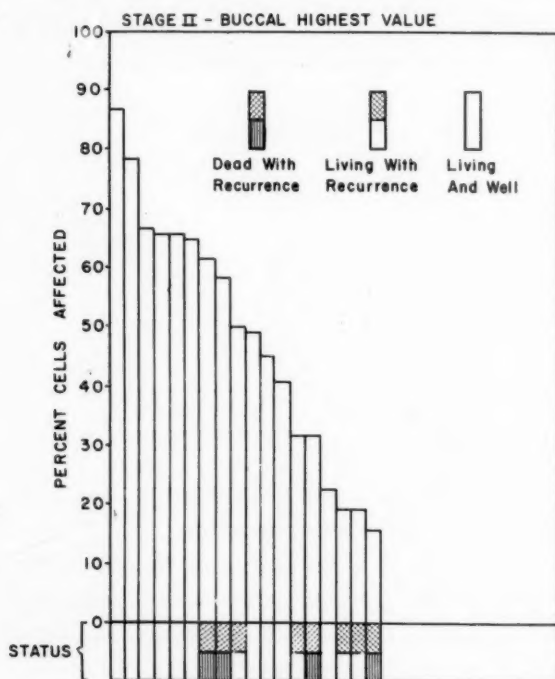


Fig. 7.—Buccal smear. Highest radiation response and 2 year end result in 18 patients with Stage II carcinoma of cervix.

This experience may be contrasted with the latest series of Graham,¹⁸ who reported 31 Stage I patients. Of these, 17 had good response and 11 (65 per cent) were living and well at the end of 5 years. There were 14 patients with poor response and 2 (14 per cent) were living and well at the end of 5 years. It is to be noted that the Graham group was apparently subject to a high degree of selection and this makes comparisons difficult. Thus, of the 31 Stage I cases of both good and poor radiation response, there were only 13



Fig. 8.—Photograph of projection apparatus used to measure cell area.

survivors for a 5 year cure rate of but 42 per cent. With a more conventional Stage I cure rate a test of great precision would be required to segregate the potentially curable from the noncurable. It must be noted, however, that Kjellgren's¹³ careful study confirms the prognostic value of RR in an unselected Stage I series.

There were 33 patients with Stage II carcinoma of the cervix available for analysis. Of these, 15 had a high radiation response and 8 or 54 per cent had no evidence of recurrence after 2 years. There were 18 patients with low radiation response and 8 or 44 per cent had no evidence of recurrence at the same period of time (Fig. 2).

The Stage II figures may be contrasted with the Graham series of 73 patients. In that series 24 of 35 (69 per cent) high response patients seemed well while 5 of 38 (13 per cent) low response patients lived. This difference is highly significant and the over-all cure rate is conventional for this stage.

Estimation of Radiocurability by 4 Morphological Characteristics of Buccal Cells After Standard Test Irradiation

The standard test irradiation consisted of 1,500 r administered through a 3 cm. cone to the right buccal mucosa just inside the angle of the mouth. The factors were: 100 kv., 5 Ma., 15 cm. STD, 0.5 mm. aluminum filter. Patients received an antibiotic troche* to use during the height of the reaction. Smears were obtained 3 times a week for 3 weeks and prepared by the method of Papanicolaou. Slides so obtained were relatively free of debris, were much easier to read than similar slides from the vagina, and contained only normal epithelial cells and leukocytes which appeared about 10 days after radiation. The cell count was by the same 4 criteria (Fig. 3) used in the Graham vaginal cell technique except that the critical level of response was considered to be above 65 per cent rather than above 75 per cent. The majority of patients



Fig. 9.—The Ott planimeter in position to measure cell area. Note the shadow pointer which greatly facilitated the procedure.

were tested retrospectively, i.e., when they had recurrences or when they had survived for more than 5 years. The series is therefore not consecutive, so that the end results cannot be representative for the stage. Typical response curves are seen in Figs. 4 and 5.

There are available for analysis 29 patients with Stage I carcinoma. There were 14 patients who had greater than 65 per cent response and all of these patients were without evidence of disease after 2 years. There were 15 patients whose response was below 65 per cent and, of these, 5 had recurrence within 2 years (Fig. 6).

There were 18 patients with Stage II carcinoma. Only 3 of these had a response above 65 per cent and all of these were well after 2 years. Fifteen patients had a response below the critical level and 8 of these had recurrence within the 2 year period (Fig. 7).

*Spectrocin-T, supplied by E. R. Squibb & Sons.

If the Stage I and Stage II cases be considered together, there were 17 patients whose response was above the critical level and all these were well. There were 30 patients below the critical level and 17 of these were well and 13 had recurrences within the 2 year period.

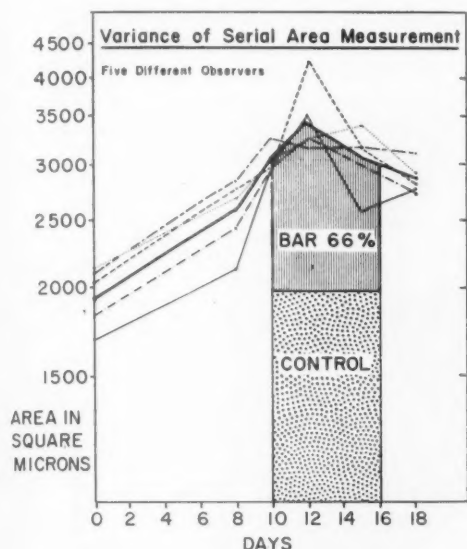


Fig. 10.

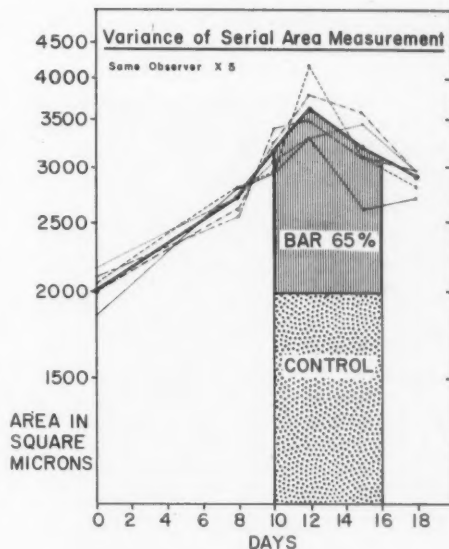


Fig. 11.

Fig. 10.—Variation in area measurement by 5 different inexperienced observers with the Ott planimeter. The buccal area response (BAR) is calculated as the mean increase in cell area during the week of maximal response.

Fig. 11.—Variation in area measurement by the same observer, using the same slides measured in Fig. 10.

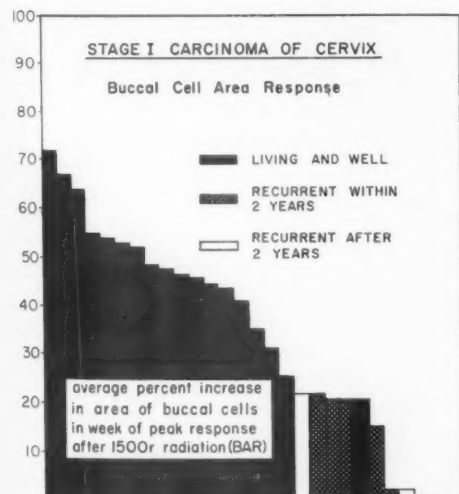


Fig. 12.

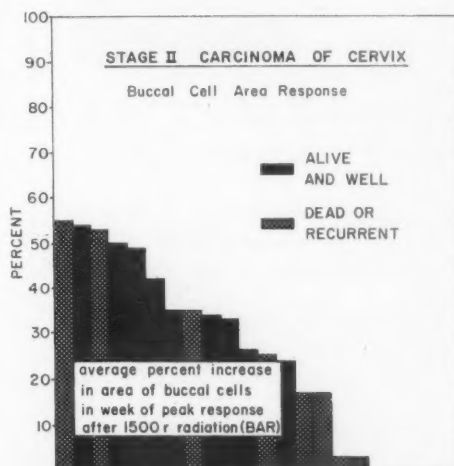


Fig. 13.

Fig. 12.—The buccal cell area response according to 2 year end result in 25 patients with Stage I carcinoma of the cervix.

Fig. 13.—The buccal cell area response according to 2 year end result in 17 patients with Stage II carcinoma of the cervix.

Estimation of Radiocurability by the Buccal Cell Area Response After Test Irradiation

In an attempt to be more precise by the elimination of observer bias, the buccal cells were subjected to analysis for increase in cell area. This was determined directly from microprojected images with an Ott planimeter having

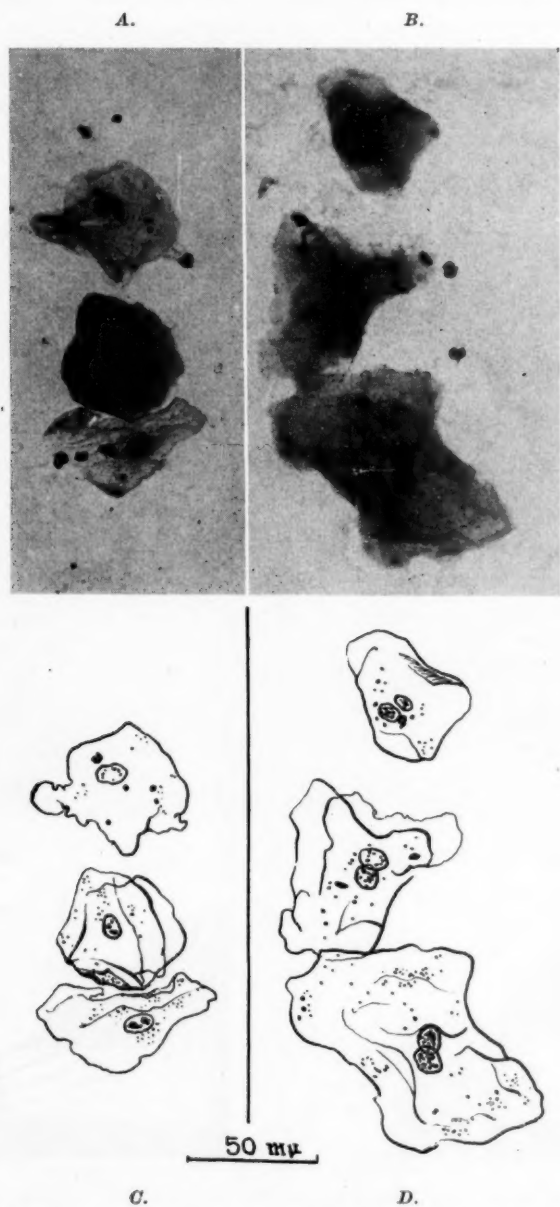


Fig. 14.—A, A 12 day smear from an unresponsive patient. There is so little change from the control (Fig. 3, A) that it may be used as a comparison for the cells in B. ($\times 500$; reduced $\frac{1}{2}$.) B, A 12 day smear showing multinucleation and some cell enlargement. ($\times 500$; reduced $\frac{1}{2}$.) C and D, Pen and ink sketch emphasizing cellular features.

a shadow pointer on the tracer arm (Figs. 8 and 9). Fifty cells were measured on each slide. The projector height was adjusted to produce $\times 1,000$ magnification and the planimeter was calibrated to read directly in square microns. Fifty cells per slide but only one cell per field were measured; an additional 100 or more cells were measured if variation was encountered.

The technique has the advantage of relative speed and reproducibility with personnel considerably less trained than for the morphological method. The area response is calculated as the mean increase in cell area over the control value in the week of maximal response. Figs. 10 and 11 show the limits of reproducibility for relatively untrained personnel as well as for a trained person and shows the method for charting and calculating the response. Further details of the method are elaborated in a separate publication.

Among the 25 Stage I cases, the correlation of buccal cell area response and clinical end result was quite good. A 25 per cent increase in area appeared to be the minimum required for a good clinical response in this stage

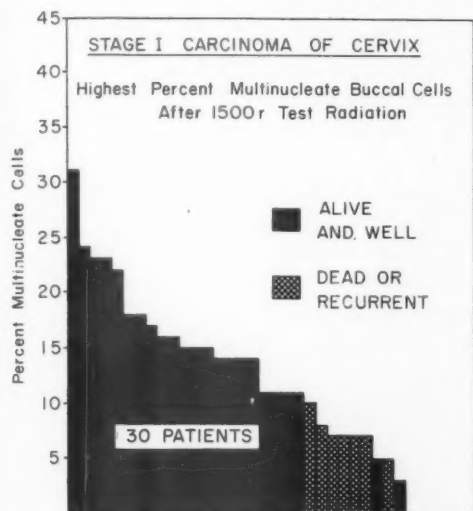


Fig. 15.

Fig. 15.—The highest per cent multinucleate buccal cells and 2 year end results in 30 patients with Stage I carcinoma of the cervix.

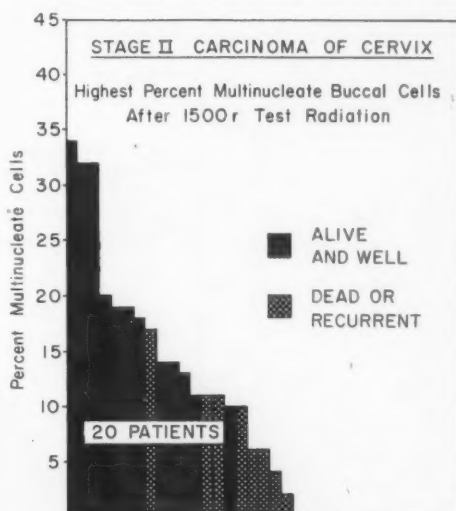


Fig. 16.

Fig. 16.—The highest per cent multinucleate buccal cells and 2 year end results in 20 patients with Stage II carcinoma of the cervix.

of carcinoma. All 17 patients with areas above the critical level were well. Of 8 patients below the critical area response, 5 had recurrences within 2 years of initial therapy and 2 others were retrospectively treated patients who had recurrences after 2 years (Fig. 12).

Only 17 Stage II cases were available for analysis. Although there is a tendency for the recurrent cases to be among the patients with low area response, the division is not so clear-cut as in Stage I (Fig. 13).

Estimation of Radiocurability by the Percentage of Multinucleation of Buccal Cells After Test Irradiation

The determination of the per cent of multinucleated cells after test radiation is conveniently done with the high dry or oil immersion objective (Fig. 14). By use of rigid criteria for counting—only solitary cells, the first cell in a field, and only one cell per field—reproducible counts are readily obtainable.

The response is expressed as the highest per cent of multinucleated cells obtained. In the average case the highest per cent of multinucleated cells appeared between days 10 and 13 as a rather sharp peak.

The assay of multinucleation is by far the quickest method of examination. Interestingly enough, the correlation with radiocurability was strikingly good.

Thirty patients with Stage I carcinoma of the cervix were available for analysis. If 9 per cent or more exfoliated multinucleated cells be considered a good response, all patients above this critical count are well and 7 of the 9 below it have had recurrences (Fig. 15).

Twenty patients with Stage II carcinoma were available for analysis. If 12 per cent or more exfoliated multinucleated cells be considered a good response, only one patient above this figure has had a recurrence and 7 of the 9 below have (Fig. 16).

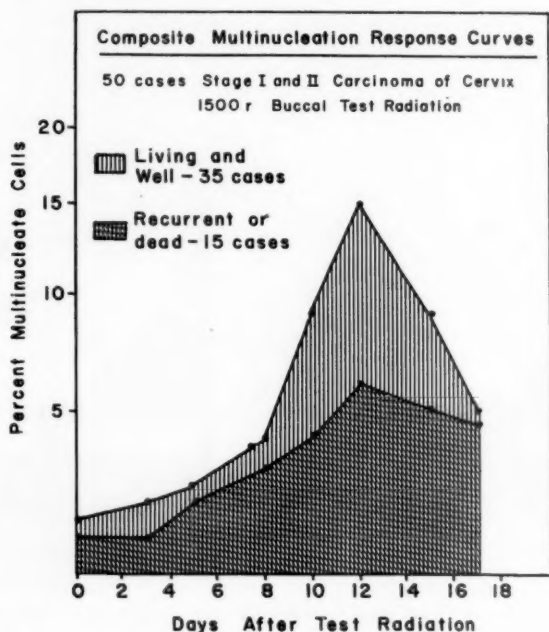


Fig. 17.—Composite multinucleation curve of survivors compared to similar curve for patients with recurrent tumor.

If the entire 50 Stage I and Stage II patients be considered as a group there is a sharp separation of the surviving patients from those with recurrences (Fig. 17).

Changes in Nuclear Desoxyribose Nucleic Acid Content of Vaginal and Buccal Cells After Irradiation

The final phase of these investigations concerns the amount of desoxyribose nucleic acid (DNA) per nucleus in the vaginal and buccal cells before and after radiation. Information on the effect of radiation in the DNA of carcinoma is sparse and in some respects conflicting. Biochemical studies have shown that there is about twice the DNA content in cervical cancer when compared to an equal weight of normal cervix. On the other hand, cytohistological studies have shown that individual cells in a tissue section

may contain greatly increased amounts of DNA although other cells, apparently dying, contain less than the diploid amount of DNA. Mellors and associates²⁰ have confirmed this in exfoliated vaginal cells and we have been able to confirm this in many cases.

Taymor and co-workers²¹ and Gusberg¹⁷ have shown, by biochemical and cytohistological methods, respectively, that the DNA content of tumor cells seems to be decreased following radiation. A careful examination of the biochemical data, however, shows that, while there is a mean decrease in DNA content, the effect in individual patients varied widely. Taymor and associates raised the question of whether these variations might be an individual expression of radiosensitivity but, unfortunately, the shortness of the follow-up precluded correlation with radiocurability. Gusberg's preliminary cytohistological findings were consistent with the thesis that a relative diminution of DNA after radiation was associated with local healing of the tumor, but radiocurability was not assayed.

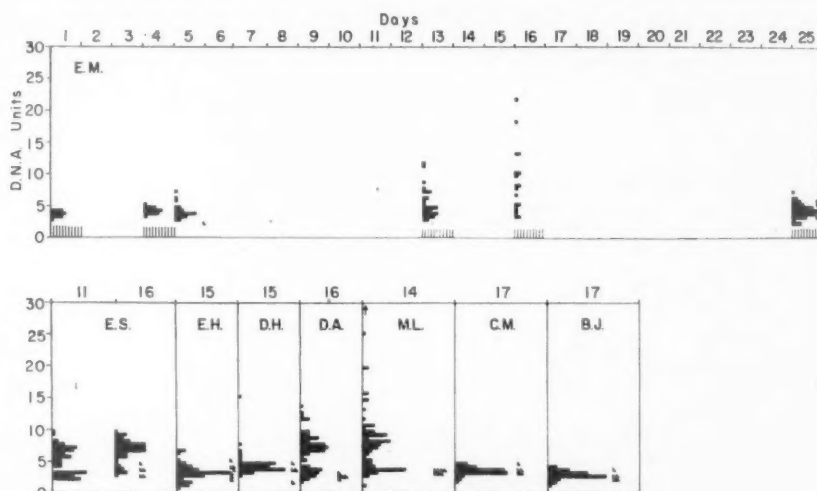


Fig. 18.—The shift in total nuclear desoxyribose nucleic acid in exfoliated buccal cells after radiation in 8 patients. Chart of E. M. shows maximal response on day 16. Only day of maximal response is shown in other 7 patients. Each small solid square represents one epithelial cell. Each small solid triangle represents a white blood cell. Patients B. A. and M. L. demonstrate maximal response while the others show very little response.

Our investigations have been directed toward examining the possibility that individual variation in the DNA content of exfoliated cells following radiation might be a measure of radiocurability.

The information gathered was obtained by microspectrophotometry according to the method of Caspersson²² as modified by Pollister and co-workers²³ and Patau.²⁴ Over 3,000 individual cell measurements from 25 patients with Stage I and Stage II carcinoma of the cervix have been determined. The slides of 11 patients after vaginal and 13 patients after buccal irradiation were examined during a period of about 4 years. Because of the small number of patients and the similarity of findings, the buccal and vaginal data will be considered together. In general, 3 DNA patterns may be distinguished in the normal cells after radiation; in one there is a minimum number of cells with nuclear DNA above the diploid value; at the other extreme there is a group where there is the accumulation of a relatively large number of exfoliated cells with DNA in excess of the diploid value; and in the third group the number of cells with greater than diploid DNA is intermediate between the

two extremes (Fig. 18). In some instances cells contained up to 16 times the normal amount of DNA. Many nuclei showed a dilution of DNA with increase in area, indicating a failure of synthesis to keep pace with increase in size, but the cytoplasmic and nuclear areas of other cells seem to be increased roughly in proportion to the increase of nuclear DNA content (Fig. 19), thus indicating continued DNA synthesis while growing.

If the small number of patients available for analysis is tabulated according to the DNA behavior and the clinical end result, it is evident that the 5 patients who exfoliated a large number of cells with increased DNA survived without evidence of recurrence for at least 2 years. On the other hand, of 15 patients with a minimal number of cells with increased DNA, 9 showed evidence of recurrence within 2 years. The 4 intermediate cases represented 2 patients who survived and 2 who had recurrence (Table I).

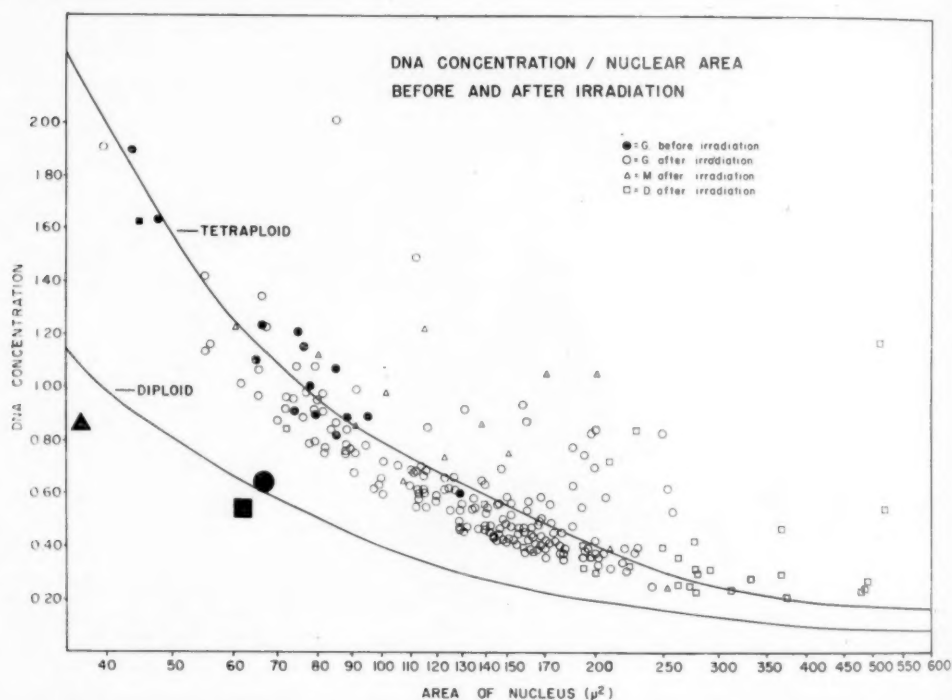


Fig. 19.—The relation between desoxyribose nucleic acid and nuclear area. Solid symbols—before radiation; hollow symbols—after radiation. Large symbols—mean value; small symbols—individual cell value.

TABLE I. DNA ACCUMULATION CORRELATED WITH 2 YEAR CLINICAL STATUS OF 24 PATIENTS

DNA ACCUMULATION	WELL	RECURRENCE
Minimum	6	9
Intermediate	2	2
Maximum	5	0

Although the DNA data are treacherous to interpret because of the small number of patients, in spite of 4 years' work, it is helpful to have a working hypothesis which at least is consistent with the data. The accumulation of cells with greater than diploid amounts of DNA is probably an expression of the inhibitory effect of x-rays on mitotic division. This may mean that, in the

cells in question, the inhibition of mitosis is not necessarily accompanied by a proportional inhibition of DNA synthesis or by the other enzyme systems which have to do with cell growth, at least in the dose administered. This phenomenon has been observed in cells in tissue culture.²⁵ It is only possible to speculate on whether or not the ability of x-rays to preferentially inhibit the mitotic process and thereby form giant cells is a measure of the elusive property of radiosensitivity. The data are at least not against this view.

Significance of a 2 Year Follow-up Period

Lombard²⁶ has recently studied the 2 year and 5 year survival rates over a number of years for carcinoma of the cervix in 3 important Massachusetts hospitals. She found that there was an improving survival rate for patients treated for cancer of the cervix for the period 1927 through 1952. Furthermore, it appeared that the 2 year survival rate exhibited the same trend as

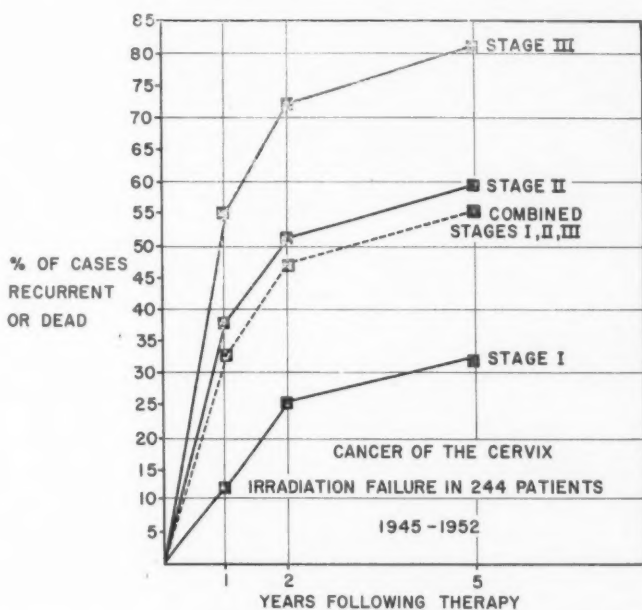


Fig. 20.—Recurrence rate by years. Irradiation failures only, without consideration to subsequent surgical cure.

the 5 year rate except that it centered at a somewhat higher level. She concluded that 2 year survival rates gave a satisfactory basis for comparing methods of treatment. In the case of the Massachusetts General Hospital, there was a 15 per cent difference between the 2 and 5 year rates.

We have compared the 2 and 5 year radiocurability rates in our own material for the period 1945 to 1952, inclusive. Only Stage I, II, and III cases are included. No case treated primarily by operation is included. The 5 year radiocurability figures do not correspond to the clinical 5 year survival figures, because, for our purposes, any patient found to have a radioresistant tumor and secondarily operated upon and thereby cured is, nevertheless, considered as having had a recurrence. Furthermore, the few unfollowed cases are excluded. Examination of the data by stages reveals the fact that a great majority of radiation failures were apparent by the end of 2 years regardless of stage (Fig. 20, Table II).

TABLE II. APPARENT CURE RATE AFTER 1, 2, AND 5 YEARS BY RADIATION THERAPY (1948-1952)

STAGE	NO. OF PATIENTS	APPARENTLY CURED					
		1 YEAR		2 YEARS		5 YEARS	
		NO.	%	NO.	%	NO.	%
I	84	74	88	63	75	57	68
II	107	67	62	52	49	44	41
III	53	24	45	15	28	10	19
Total	244		67		53		45

The use of 2 year radioresistant figures thus introduced a maximum error of 7 per cent as compared with 5 year statistics for Stage I and 8 per cent for Stage II.

Summary

An effort was made to correlate the 2 year clinical end results with radiation changes in exfoliated vaginal cells according to the Graham technique. Unfortunately, in our hands, this was not successful.

The concept of generalized host radiosensitivity was examined by administration of a test dose of irradiation to the buccal mucous membrane after which certain changes in the exfoliated cells were observed. Two features of radiation cytotoxicity—macrocytosis and multinucleation—proved to lend themselves well to objective measurement and correlated in high degree with the clinical end results in the retrospective study of 50 Stage I and Stage II carcinomas of the cervix. With multinucleation as the sole criterion, the prognostic error was but 12 per cent in these operable patients. At the present time a prospective study is in progress.

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Discussion

DR. WILLIAM F. MENGERT, Chicago, Ill.—The search for some reliable method of predicting how the cancer cells of a given patient will respond to irradiation continues. In my early days in this honored specialty, attention was focused on the degree of anaplasia of the cancer cells. Later it was decided that glandular types of tumor did not respond as well to irradiation as did squamous types. Therefore, Wertheim's operation was often performed for adenocarcinomas of the uterine cervix. Finally, the obstetric and gynecologic world arrived at the simple truth that the all-important prognostic characteristics of any cervical cancer were its size and extent beyond the site of origin at the time treatment is begun.

Nevertheless, attempts continue to be made to correlate various characteristics and properties of the cell with ultimate patient survival. These attempts are important because they add to our knowledge of cellular biology, from which will come the ultimate solution of the problem of malignant disease. Certainly others, including the experts at my institution, after prolonged and exhaustive attempts, cannot reproduce the experience of the Grahams. Dr. Jones recognizes this, and in his paper states that "it seems clear that there is difficulty in easily transferring the method to other laboratories."

Dr. Jones and his associates have hit upon an ingenious idea, namely, study of the response to irradiation of mucous membrane cells of the same individual, but at a site considerably removed from the original tumor. They have studied 50 patients in retrospect. This number of patients is not statistically significant; therefore, I hope this work will continue. Studies of cancer necessarily involve large numbers of patients over many years before firm results begin to emerge.

The director of any large clinical department of obstetrics and gynecology must make a continuing series of choices concerning research to be pursued, and often with only meager knowledge. Some lines of endeavor seem to offer promise in the peculiar circumstances of personnel and material of that department; others do not. In such light we examined the work of the Grahams—and Dr. Jones's work cannot be considered separately from this—and decided that our opportunities did not lead in this direction.

We will watch with interest as Dr. Jones pursues these researches, and will look forward to the time when he is able to give us his conclusions concerning them.

DR. HERBERT E. SCHMITZ, Chicago, Ill.—Three years ago we reported before the American Association our experience with basal cell counts to determine radiation response. Not being able to duplicate all the study of the Grahams, we noticed in our smears that we had a greater number of basal cells in response to a known dosage of radiation. At that time we reported 35 cases with similar results to these studies of the cells in the buccal mucosa. We have in the meantime added to those and have 100 such cases, carefully studied. At first we were surprised to see these patients live into the second and third years, having shown very poor response at the first study. Since that time we recognized the fact that response to radiation is not immediate. In going back over the charts of the patients that are living, we found that at the end of 30 days some had satisfactory response, and others showed this still later. I am quite sure that Dr. Jones will find in his study a similar experience, and I would like very much to carry this on as a comparison between the 2 methods.

DR. ABRAHAM E. RAKOFF, Philadelphia, Pa.—I think many laboratories throughout the world have been anxiously following the results of the concept of the Grahams on the significance of SR and the significance of RR. The present paper deals with RR. We have had the opportunity over the years to follow patients before and during radiation with regard to both SR and RR. Several years ago 2 of my associates and I undertook to do quantitative counts on cells from patients who had had radiation for malignant disease of the pelvis. We noticed, as did the Grahams, that there was a remarkable difference in different patients, so far as response of the vaginal cells was concerned, with regard to

enlargement of the cell, with regard to vacuolization, with regard to multinucleation, and particularly with regard to enlargement of the nucleus as well as to the distribution of chromatin. Although we have not yet published our figures, we do believe that there has been a positive correlation in the retrospective cases with regard to RR and the effects of radiation therapy.

I think the important lesson I have learned from Dr. Jones's work is the necessity for objectively quantitating the various factors that the Grahams have mentioned. Any technician who reads RR knows how difficult it is to quantitate these various factors, so it is important to follow this lead and actually measure the area of enlargement of the cells and the multinucleation. We were much impressed in our observations with the enlargement of the size of the nuclei, and I would like to ask Dr. Jones whether he undertook nuclear measurements as compared to the size of the cell as an objective indicator of his findings.

DR. SAUL B. GUSBERG, New York, N. Y.—As a person who has battered and bloodied my head against this problem of radiosensitivity in attempting to analyze not only my own work but that of others, I welcome the clean end point that Dr. Jones has outlined in his program. I think it will permit him to obtain an evaluation of the method very quickly but it will also introduce something new into this idea of testing: it will enable us to evaluate his work very quickly. I am disturbed about the 1,500 r dose to the normal buccal mucosa, but perhaps that will not prove harmful.

DR. JONES (Closing).—The matter of SR was mentioned. We have not included SR in the paper but we do have SR data. These data, like those in Dr. Gusberg's recently published experiences, do not correlate with the end results. I think it is wrong to say that everyone who has tried the Graham technique cannot do it. Dr. Schmitz mentioned a modification which proved helpful. Dr. Rakoff likewise. There is a very excellent report by Kjellgren that I think must be considered to be confirmatory in every respect, so that we are prepared to say that for the reasons I mentioned, one must pay very close attention to the fine print in the Graham technique.

We have not measured the nuclear size in a large number of patients. We have measured it in connection with some DNA measurements of exfoliated cells and I suspect that one could get the same result with nuclear enlargement as with whole cell enlargement.

While collecting these data, we had an interesting experience. We selected one patient who had been well for 5 years for retrospective study. She was tested with the buccal technique and Dr. Davis measured her cells and said that we had a patient who did not respond and who was perfectly well. Several of us measured this patient's cells and none of us could get a response. We went back over her history and found that when she was originally treated, there was discussion as to whether she had Stage I carcinoma or an intraepithelial lesion. Finally, it was decided that she had Stage I carcinoma and, therefore, she was treated by irradiation. The physician who administered the radium thought he would take one final series of biopsy specimens, and in this last effort no tumor was found. Therefore, it was an exceedingly early case. After the buccal test, the patient was called back to the clinic, and on examination the cervix seemed to be all right but it was biopsied, and we were interested and amazed to find that the cervix was riddled with carcinoma. This aroused our interest a great deal and, although one robin does not make a spring, we were stimulated by the experience.

TWO CASES OF SUPERNUMERARY OVARY AND ONE OF ACCESSORY OVARY, WITH AN ANALYSIS OF PREVIOUSLY REPORTED CASES*

LAWRENCE R. WHARTON, BALTIMORE, MD.

(From the Department of Gynecology of the Johns Hopkins University and Hospital)

SUPERNUMERARY ovary is one of the rarest of gynecological conditions. There has been only one recognized case among all the patients who have been admitted to the Johns Hopkins Hospital, and only one seen in the longer history of the Hospital for the Women of Maryland. Likewise, there has been only one case of accessory ovary among 93,537 admissions to the Gynecological Service of the Johns Hopkins Hospital. This rarity is emphasized by the fact that, in the entire medical literature of the world, there have been only 2 short articles on this subject since 1931. For this reason, it seemed worthwhile to record the rather unique case of supernumerary ovary that came under my care recently and to include the single cases of supernumerary and accessory ovary from the records of the Johns Hopkins Hospital. At the same time, I have taken the opportunity to study and assemble the available records of all previously reported cases.

Supernumerary Ovary.—

CASE 1.—History No. 406,831, Hospital for the Women of Maryland. I first saw this woman in 1949, when she was 30 years old. She was white, married, had never been pregnant, and had always had excellent health; she was well developed and intelligent. In 1949 she had a very mild chronic salpingitis which needed only conservative care. I saw her once every 6 months during the next 7 years, during which time she had no significant symptoms.

In September, 1956, because of increasing pain, intermenstrual bleeding, and increase in size of the ovaries, I operated upon her. There was a bilateral chronic salpingitis with hydrosalpinx. The ovaries were cystic, measuring about 7 cm. in diameter; the uterus was normal, except that it was densely bound in the adnexal adhesions. I performed a total abdominal hysterectomy, bilateral salpingo-oophorectomy, and appendectomy.

After the pelvic organs were removed and before the pelvic operative field was peritonized, I explored the pelvis, as usual, and found what seemed to be an enlarged lymph gland, entirely separate from the pelvic organs, far out on the right pelvic wall beyond the right ureter. It was 2.5 cm. long and about 1.5 cm. thick. I removed it. Microscopic examination of this structure proved that it consisted entirely of ovarian tissue, which, in addition to the 2 normally placed ovaries, made a total of 3 ovaries (Figs. 1 to 3).

The pathological diagnosis of the other pelvic organs was chronic salpingitis, bilateral; serous cysts of ovaries, bilateral, with early endometriosis. The uterus was normal.

The hospital convalescence was normal. A routine postoperative examination, one month after discharge, revealed normal findings. The postoperative hot flushes lasted about one month.

*Presented before the Eighty-second Annual Meeting of the American Gynecological Society, Hot Springs, Va., May 21-23, 1959.



Fig. 1.

Fig. 1.—Ovary No. 1. Shows normal ovarian stroma, atretic follicles, corpora albicantia.

Fig. 2.

Fig. 2.—Ovary No. 2. Shows wall of a fairly recent corpus luteum and an old hyalinized corpus, with normal ovarian stroma.

Fig. 3.

Fig. 3.—Ovary No. 3. Supernumerary ovary. Shows wall of a corpus luteum, with stroma in base.

In December, 1957, 15 months after the first operation, I palpated an asymptomatic mass, 8 cm. in diameter, apparently about the level of the promontory of the sacrum and about 5 cm. to the left of it. It had no connection with the pelvis and was apparently retroperitoneal. By Jan. 27, 1958, it had become larger, about 10 or 11 cm. in diameter, and I advised its removal. This was done on February 21, 1958, at the Hospital for the Women of Maryland. Since I suspected that it might involve the sigmoid and possibly necessitate intestinal operation, I asked one of my associates, Dr. I. Ridgeway Trimble, Associate Professor of Surgery at the Johns Hopkins Medical School, to be present and help, if necessary. Before performing the operation, because of the unusual history and possibilities, I performed cystoscopic and gastrointestinal studies, bilateral pyeloureterograms, cystogram, barium enema, and gastrointestinal roentgenograms, all of which revealed no abnormalities of the gastrointestinal or urinary organs.

At operation, I found the mass to be located in the retroperitoneal tissues, in the base of the mesentery of the sigmoid. It was about 10 cm. in diameter. The median half consisted

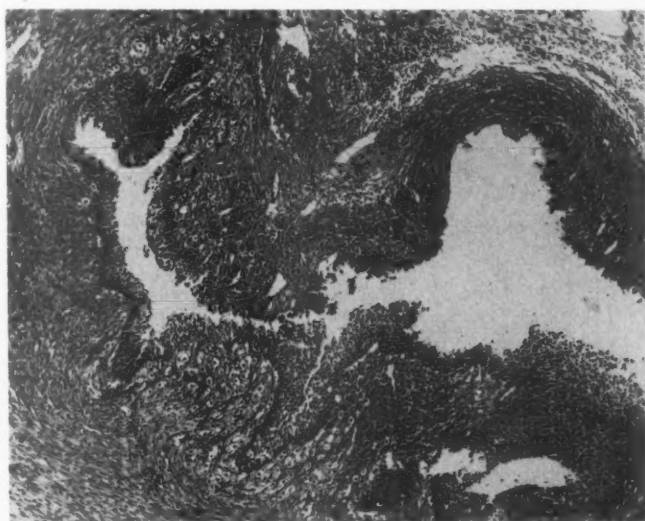


Fig. 4.—Ovary No. 4. Supernumerary ovary. Shows wall of a fresh corpus luteum, with normal ovarian stroma.

of a thin-walled cyst containing clear fluid. The lateral half consisted of a soft mass which looked exactly like lobules of corpus luteum tissue. Microscopic examination established the diagnosis of ovarian tissue, confirming the fact that this was ovary No. 4. Again, there were a few hot flushes which disappeared after a month or so (Figs. 4 and 5).

After a normal convalescence, 2 months after the second operation, this patient developed a fifth mass, of the same size, and shape, and in the same location as ovary No. 4. It was round, smooth, cystic, and about as large as a man's fist, located just to the left of the sacral promontory. I advised observation, and within 6 weeks this mass disappeared completely and has not returned.

In June, 1958, 4 months after the second operation, a sixth mass appeared in the right half of the pelvis, about as large as a man's fist, having all the characteristics of a simple, clearly outlined cyst. This mass was in the same location as ovary No. 3. It also disappeared spontaneously and subsequent examinations at intervals of 3 months have revealed no further sign of ovarian activity or pelvic masses.

This is therefore a proved case of supernumerary ovary, consisting of 4 ovaries. The 2 supernumerary ovaries were completely separate from the 2 normally placed ovaries. One was on the right pelvic wall, extraperitoneal. The fourth was in the base of the mesentery of the sigmoid, retroperitoneal. These diagnoses were all confirmed microscopically. The

2 subsequent cystic structures which formed and disappeared spontaneously were possibly follicular cysts developing in ovarian tissue in those areas. That diagnosis, however, is conjectural and was not confirmed microscopically.

CASE 2.—History No. 756,296, Johns Hopkins Hospital; Autopsy No. 26,984. This was a 20-year-old white woman, married 3 years, who had had 2 spontaneous abortions. The interval between the 2 miscarriages was only a few months, the last one being in November, 1955. This was followed by severe continual bleeding, for which a curettage was done in Covington, Virginia. A diagnosis of chorionepithelioma was made and a total abdominal hysterectomy with bilateral salpingo-oophorectomy was performed on May 25, 1956, in the Chesapeake and Ohio Railroad Hospital, Clifton Forge, Virginia. In August, 1956, because of cerebral metastases, a decompression was performed, apparently in Roanoke, Virginia, with temporary relief. In November, 1956, she was first admitted to the Johns Hopkins Hospital and given palliative therapy; she was discharged on December 8 and readmitted on Dec. 29, 1956. She died the following day.

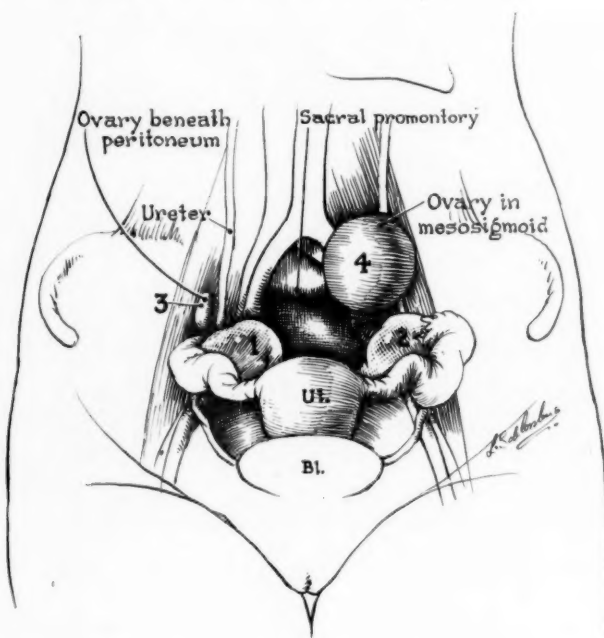


Fig. 5.—A drawing showing the locations of the 4 ovaries removed from patient in Case 1 and proved by microscopic examination.

At autopsy, the pathological diagnosis was metastatic chorionepithelioma, agenesis of the left kidney, left ureter, left renal artery and vein, surgical absence of the uterus, tubes, and ovaries, and ectopic ovary. The ectopic ovary was an irregular mass of tissue, 2 by 2 by 2.5 cm., situated to the left of the aorta. The pathologist who did the autopsy first said it might be a lymph node, judging from its gross appearance. It consisted of ovarian tissue.

This diagnosis was not questioned until the spring of 1959, when Dr. Arnold Rich, the Emeritus Professor of Pathology, volunteered to review all the records of his department for me, to see whether they had ever had a case of supernumerary ovary. As soon as he saw the record of this case, he realized that this would be a case of supernumerary ovary if it could be proved that the patient had had 2 ovaries previously. A letter to the Clifton Forge Hospital brought the information that the operation performed there consisted of abdominal hysterectomy and bilateral salpingo-oophorectomy and that the operative specimen contained 2 normal ovaries. This was therefore a second authentic case of supernumerary ovary, with

the associated congenital malformation of agenesis of the left urinary tract (Fig. 6). This is the only case of supernumerary ovary in approximately 29,000 autopsies, and the only case in the clinical records of the hospital.

Accessory Ovary.—

CASE 3.—Gynecology Service No. 38,211, Johns Hopkins Hospital; Gynecologic Pathology No. 37,230. The patient was a 21-year-old Negro woman, admitted to the Johns Hopkins Hospital, July 20, 1931. The essential clinical diagnosis was chronic salpingitis, and the essential operative procedure was right salpingectomy. The operator, Dr. Hugh Warren, did not see the tiny structure, 4 mm. in diameter, attached to the mesosalpinx near the fimbria of the right Fallopian tube. The pathologist, Dr. Herman Long, however, saw it and said he thought it was a lymph gland. Microscopic section showed that it contained numerous small follicles, one corpus albicans, and some small ova (Fig. 7). The diagnosis of accessory ovary was made.

Review of the Literature

In 1861, in the third edition of his *Lehrbuch der Pathologischen Anatomie*, Carl Rokitansky stated that no case of supernumerary ovary had yet been seen.

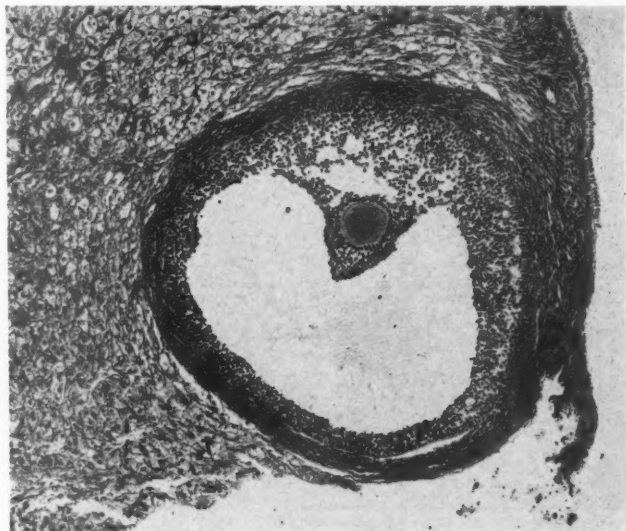


Fig. 6.—Case 2. Supernumerary ovary. Shows typical ovarian follicle, stroma, and lutein tissue. The only case on record in the Johns Hopkins Hospital and in approximately 29,000 autopsies.

The same opinion was expressed by Klob in 1864 in his textbook, *Pathological Anatomy of the Female Sexual Organs*. In 1863, Grohe exhibited the first recorded case of 3 ovaries before the Clinic at Stettin. This case he published formally in 1864.

Since that date, 54 authors have published articles reporting various types of supernumerary or accessory ovary; in addition, various studies have been published in which no new cases have been reported. I have personally read and abstracted all but 9 of these articles; these 9 could not be found. Some of these reports are long and studious articles of many pages; others are simply case reports not more than 10 or 12 lines in length, without any pathological confirmation.

Obviously, one of the first problems in analyzing this material is to establish a satisfactory classification. Since this is basically a problem in embryology and pathology, I consulted the members of the Department of Embryology of the Carnegie Institute of Washington and the Department of Pathology.

In practically all serious studies of this subject, since the first report by Grohe in 1864, various authors have attempted to classify these anomalies. There has been general agreement on 3 basic principles:

1. The term "supernumerary ovary" (*ueberzachliges ovarium*, *ovaire surnuméraire*) includes those rare cases in which the third ovary is entirely separate from the normally placed ovary and apparently arises from a separate primordium or anlage.

2. The term "accessory ovary" includes those cases in which the excess ovarian tissue is situated near the normally placed ovary, may be connected with it, and seems to have been developed from it, possibly from tissue that was split from the embryonic ovary during its early development. It is invariably located near the normally placed ovary.

3. The supernumerary or accessory ovary must contain ovarian follicular tissue.

These 3 criteria are the basis on which I have attempted to classify these cases.

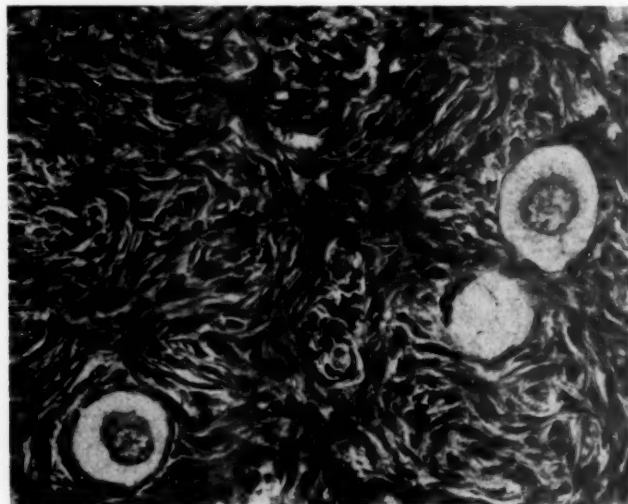


Fig. 7.—Accessory ovary. Shows typical follicle and ovarian stroma. The only accessory ovary on record in the Johns Hopkins Hospital.

During the past years, various additional criteria have been added, some of which are arbitrary, have no embryological foundation, and have produced only clinical confusion. These specifications have been suggested chiefly to distinguish supernumerary from accessory ovaries. Among these criteria are the following:

1. The supernumerary ovary must have normal ligamentary connections with the uterus, Fallopian tubes, or ovaries.

2. The supernumerary ovary must be of a certain size and must have reached a certain stage of development, resembling the normal ovary in shape and size.

3. The supernumerary ovary must be separated from the normal ovary by a specified number of centimeters to distinguish it from an accessory ovary.

4. If the excess ovarian tissue is attached to the anterior surface of the broad ligament, it is a supernumerary ovary.

For self-evident reasons, there could be no agreement about some of these arbitrary criteria, such as the required size, shape, or exact distance from the

normal ovaries. Furthermore, the clearest cases of supernumerary ovary are those that have absolutely no connection with any of the pelvic organs and develop in retroperitoneal regions.

In classifying these cases, another finding has been emphasized, namely, the presence of associated congenital malformations of the genitourinary organs. Thus, in 3 of the 4 cases of supernumerary ovary and in about 26 per cent of the cases of accessory ovary, additional defects have been found. These associated malformations have been accessory Fallopian tube, accessory tubal ostium, bicornuate or unicornuate uterus, septate uterus, agenesis of the kidney and ureter, diverticulum of the bladder, and anomalies in other organs. These findings indicate that there has been a primordial defect in most of the cases of supernumerary ovary, whereas such evidence is much less frequent in cases of accessory ovary. This characteristic assists in classification, but also suggests that no classification can be completely rigid and that it is impossible to be absolutely arbitrary as some cases share some of the features of both types of ovarian anomaly.

In addition to the two groups, supernumerary and accessory ovary, there are the groups of lobulated ovary; pelvic tumors apparently of ovarian origin in cases in which there are in addition 2 normally placed ovaries; and cases in which, following bilateral oophorectomy, there is a continuation of ovarian function as shown by persistence of menstruation or subsequent development of pregnancy. These groups will be discussed in order.

Supernumerary Ovary

By definition, this group includes only those cases in which the structure contains typical ovarian tissue and in which the supernumerary ovary has no connection whatever with the normally placed ovary. The second criterion leaves some room for judgment and possibly arbitrary decision. In the past, it has proved only confusing to attempt to specify the number of centimeters that should separate the supernumerary ovary from the normally placed organ. To avoid such confusion, we have excluded all cases in which there is any ligamentous or direct connection whatever with the normally placed ovary, the broad, uteroovarian or infundibulopelvic ligaments. This may seem arbitrary, but as a matter of fact, practically all cases of accessory ovary are situated so near to the normal ovary that there has been little room for doubt.

Incidence.—Supernumerary ovary is one of the rarest of gynecological disorders. Thus, in 1924, Kermauner stated that he could find no authentic case in the literature. No careful observer has listed more than 3 or 4. As far as we know, in no case has the correct diagnosis been made or even suspected before operation, and in some the diagnosis has not been made even in the operating room or at autopsy, until a later review was made.

We have reported 2 authentic cases of supernumerary ovary earlier in this paper. In addition, we have found 2 cases in the literature, reported by F. Winkel and E. Falk. Abstracts of these last 2 cases will now be presented. In 1896, Engström, and since then, others, accepted these 2 as authentic cases of supernumerary ovary.

CASE 1.—Winkel, in 1890, described the case of a 77-year-old woman who had never been pregnant, "in spite of the fact that she had 3 ovaries." She had died of cirrhosis of the liver.

At autopsy, the uterus, tubes, and ovaries were perfectly normal, in their usual situations. Attached to the anterior wall of the uterus by a band 1.7 mm. long was a third ovary, larger than the others. Its exact size was not given. The band was inserted on the anterior uterine wall, about 1 cm. anterior to the origin of the right Fallopian tube. The third ovary lay in a concavity of the posterior wall of the bladder, a diverticulum.

Attached to the posterior surface of the third ovary was a slender band which connected with a similar band, apparently a rudimentary third Fallopian tube. This continued and was attached 1 cm. from the distal end of the right Fallopian tube. The diagnosis of a third ovary was confirmed microscopically. Because of its independent position and the 2 accompanying malformations, diverticulum of the bladder and accessory Fallopian tube, it is assumed that this was a true supernumerary ovary, which probably arose from an independent primordium and not as the result of a separation from the preformed ovary (Fig. 8).

CASE 2.—In 1891, Falk reported the second authentic case of supernumerary ovary. The patient was a 37-year-old para 0, who was operated upon for recurring pelvic pain and menstrual irregularities. Both tubes and ovaries were normal except for small follicular cysts. The uterus also contained a small myoma. There was a cystic mass in the pelvis, measuring 7 cm. by 5 cm., which had no connection whatever with the reproductive organs. Its only connection was with the omentum, from which it was easily separated. The operation was bilateral salpingo-oophorectomy and removal of the cyst.

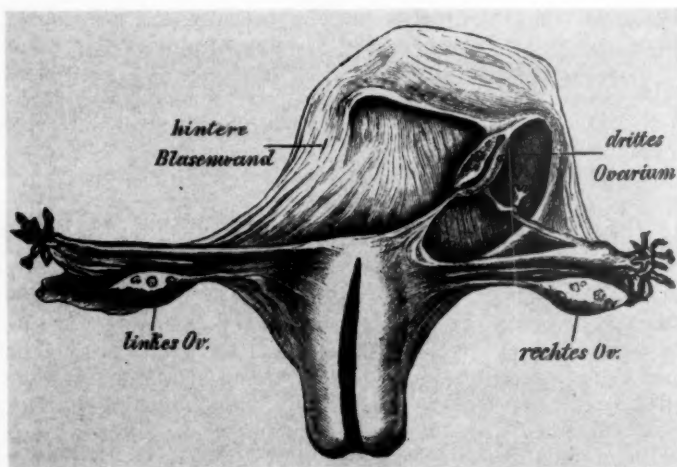


Fig. 8.—The first case of supernumerary ovary, described by F. Winckel in 1890. It is accompanied by an accessory, rudimentary, right Fallopian tube. It lay in a diverticulum of the bladder. (Courtesy of S. Hirzel, Leipzig.)

Our interest concerns chiefly the cystic mass. The 2 normally placed ovaries and tubes had no unusual features. The cystic mass consisted of a hydrosalpinx, both ends of which were closed. Its walls were 1 to 2 mm. thick. Over the hydrosalpinx was stretched an ovary, normal in size and appearance, which, on microscopic examination, was found to contain normal follicular tissue.

This was, therefore, an instance of true, authenticated supernumerary ovary, accompanied by a third Fallopian tube.

We are thus presenting a group of 4 cases which seem to us to be undoubted instances of supernumerary ovary. Two of these are from the literature, 2 are new cases reported here for the first time. In 3 of these 4 cases, there were associated malformations of the genitourinary system, some of which were undoubtedly primordial in origin—accessory Fallopian tube in 2 cases, agenesis of the left kidney, left ureter, and left renal artery and vein in the third. Also in one case was a diverticulum of the bladder.

Accessory Ovary

Cases of accessory ovary have been reported by 18 authors: Grohe, Waldeyer, de Sinéty, Beigel, Mangiagalli, Kocks, Keppler, Winckel, Schantz,

Engström, Voigt, Essen-Moeller, Doran, Smith and Wood, Latzko, Brady, Swynghedauw and Houcke, and McNeill. None of these authors reported more than one case each, except Waldeyer, Beigel, and Winckel. Waldeyer stated that he had seen several such cases; Beigel reported that he had found 23 cases of accessory ovary in 500 autopsies, and Winckel 18 such cases in 500 autopsies. Because of the lack of microscopic examination in some of these and because of the lack of confirmation by other observers, Beigel's and Winckel's reports have been largely discounted.

This category has two characteristics: the structure contains ovarian tissue and it is invariably situated near or connected with the normally placed ovary. Generally, these bits of ovarian tissue are small, almost always less than 1 cm. in diameter.

In the case we reported, the accessory ovary was so small, 4 mm. in diameter, that the operator failed to see it at all. It was discovered in the pathological laboratory, where it was first called a lymph gland.

Most frequently, they have been found attached to the broad ligament near the normal ovary. They have been described near the cornu of the uterus and between the leaves of the broad ligament.

These small bits of tissue have usually been mistaken for lymph glands or small fibromas. Even in the laboratory or autopsy room, this has been a common error, corrected only by the microscopic examination.

Also, they have usually been single, although some authors have reported finding several in one case. Waldeyer found 3 tiny bits of accessory ovarian tissue in a newborn child. They were attached to the ovary itself by short pedicles. De Sinéty reported finding a case in an infant that died soon after birth. The right ovary was deeply divided into 3 lobules and between the right ovary and Fallopian tube were 7 tiny sessile bodies, 2 or 3 mm. in diameter, attached to the broad ligament. Six of them were of Wolffian origin; the seventh had a pedicle 2 mm. long and consisted of normal ovarian tissue, stroma, and follicles.

Historical Data

Grohe, in 1864, was the first to report a case of supernumerary or accessory ovary. The previous year, at autopsy on a 40-year-old woman, para iii, he found 2 small ovaries on the left side. The right ovary was large but perfectly normal. One of the left ovaries was situated near the uterus, attached to it by the usual uteroovarian ligament. The second left ovary was farther away from the uterus, lying in a plication of pelvic peritoneum.

All 3 were examined by cross-section and found to be functioning. There is no note of any microscopic examination in the very brief case report of 12 lines. However, subsequent students of this problem, possibly having access to records not contained in the original report, have accepted this examination as having been done. We therefore accept this as an authentic case of accessory ovary, the first recorded in the literature. The brevity of the report and the lack of important details prevent one from accepting it as a true supernumerary ovary. The author himself stated that he believed the anomaly was not due to 2 separate anlagen, but rather to the subsequent division of the ovary.

Since Grohe's report in 1864, 17 others have reported cases which we have classified as accessory ovaries. With very few exceptions, this has also been the classification given by the authors themselves and subsequent students of the problem.

Quite probably, however, a few of the cases which we have classified as accessory ovaries may have been true supernumerary ovaries. Because of their proximity to the normally placed ovary, their small size, and their lack of development, we have classified them as accessory ovaries.

Although the number of reported cases of accessory ovary is considerably larger than those of supernumerary ovary, it is still one of the rare gynecological disorders.

Function of Supernumerary and Accessory Ovaries.—These small accessory and also the supernumerary ovaries have exhibited the functional and pathological capabilities of the normal ovary. Thus, in addition to normal function, as shown by the presence of active ovarian tissue, Swynghedauw and Houcke reported a case in which an accessory ovary was the source of an ovum which caused a tubal pregnancy, necessitating laparotomy and making possible the discovery of the accessory ovary. In some instances, tumors of various sorts have developed apparently in accessory, as well as in supernumerary and lobulated ovaries, as will be discussed in a following section.

Associated Congenital Malformations.—These have been found in only 5 of the 19 cases of accessory ovary. These were as follows: accessory Fallopian tube, 2; bifid Fallopian tube, 1; accessory tubal ostium, 1; accessory adrenal gland, 1; lobulated liver, 1; septate uterus, 1. This is an incidence of about 26 per cent, which is probably much higher than one would find in normal persons. It is, however, definitely lower than the incidence found in cases of supernumerary ovary.

This suggests that in some cases of accessory ovary, there is probably some defect in the germ plasm or a primordial fault in the urogenital ridge, just as seemed to be probable in the group of cases of supernumerary ovary. In that particular, this implies that there may be an embryological similarity. This explains the difficulty of classifying some cases clinically.

Clinical Characteristics of Supernumerary and Accessory Ovaries

Incidence.—It is impossible to determine the incidence of these anomalies accurately by any clinical means. They cause no symptoms whatever as they carry on their normal functions; in this respect they resemble normal ovaries. Furthermore, they may be so situated (retroperitoneally, for example) that they would be missed completely in any routine gynecological operation. Also, at times they are so small that they may be lost in the midst of gross pathological lesions. Thus, for one of these reasons, in 2 of the 3 cases we are reporting, the presence of an accessory or supernumerary ovary was entirely missed in the operating room.

Autopsy statistics give probably the most accurate estimate of their incidence. But, here again, we have shown that a case may be diagnosed as ectopic ovary. In this hospital, in about 29,000 autopsies, there has been only one case of supernumerary ovary and none of accessory ovary.

Among the 93,000 patients admitted to the gynecological service of this hospital before June, 1958, there was only one case of accessory ovary and none of supernumerary ovary. This indicates that these anomalies are excessively rare and confirms the general experience of other institutions and gynecologists.

Clinical Diagnosis.—In studying the entire literature, I have failed to find a single instance in which the correct diagnosis was even suspected before operation. Furthermore, in most cases, after seeing the lesion, the operator thought it was fibrous tissue or a lymph gland. In a few cases, the structure was so small that it was not seen by the operator, but was concealed by adherent pathological masses.

Hence, the only cases of accessory or supernumerary ovary that are ever recognized are those discovered incidentally during operations for unrelated

conditions, cases in which the accessory or supernumerary ovary develops pathological, symptomatic disorders such as tumors or cystic changes, or at autopsy. Therefore, in otherwise normal women, the exact incidence of this rare condition is impossible to determine.

Lobulated Ovary

The first case of lobulated ovary was described by Julius Klob before a medical society, apparently in Vienna, in 1866. In discussing this case, Carl Rokitsansky said he had never seen such an anomaly before.

Since then, 10 authors have reported one or more cases, making a total of 14 cases. In this group, the ovary is divided by one or more fissures into 2 or more lobules. At times, these lobules are completely separate; at other times they are connected by bands of connective tissue or ovarian stroma. Generally, there are only 2 lobes, but Schantz reported an ovary divided into 3 parts and de Sinéty, one that was divided into 4. Schantz also reported the case of a newborn child, born dead, in which both ovaries were lobulated, one divided into 3 and the other into 4 parts.

Embryologically, these cases are possibly related to supernumerary and accessory ovaries in that they represent a duplication or division of ovarian tissue. Clinically, they are at times difficult to distinguish from accessory ovaries, especially when one of the lobes is much smaller than the other.

Functionally, lobulated ovaries seem to behave just as normal ovaries do. They are likewise susceptible to various pathological changes. Klebs reported an instance in which one of the lobes developed into a small, simple cyst; Galabin reported a papillary cyst, 15 cm. in diameter, that had arisen in a lobulated ovary, and Engström removed a dermoid cyst, with a twisted pedicle, as large as a child's head, which had arisen from the outer half of a lobulated ovary.

Associated Malformations.—These have not been common in cases of lobulated ovary. They have been reported in 2 of 14 cases. Schantz found a bicornuate uterus in a newborn infant that had bilateral lobulated ovaries. Chiari reported the case of an 18-year-old girl who committed suicide. Autopsy revealed a uterus unicornis, the left half of the uterus rudimentary; the left ovary was in a femoral hernia, and there was spina bifida. The right ovary was rudimentary, divided into 2 parts, 6 and 7 mm. in diameter.

Cases of lobulated ovary have been reported by Klebs, Klob, de Sinéty, Lumniczner, Chiari Schantz, Engström, Galabin, Cavalié, and Kriss. In addition, the case of Hermann is referred to at times in the literature; however, we have not been able to find the original report of his work.

Tumors in Accessory Ovaries

The literature includes 26 reports by different authors, each recording one or more cases of pelvic or abdominal tumors in women who, in addition, had 2 ovaries. Since these tumors were of types that often arise from the ovary, these authors concluded that these tumors must have arisen from a third ovary. Hence, they have concluded that their patients had 3 ovaries, one of which gave rise to the tumor in question, and have reported them as accessory or supernumerary ovaries. The pathological diagnoses of these various tumors were as follows: dermoid cyst 6, of which one was malignant; simple cyst 5; papillary cyst 3; multilocular cyst 2; cystadenoma 2; pseudomucinous cyst 1; chocolate 1; granulosa cell tumor 1; granulosa cell carcinoma 1; myosarcoma 1. In some no pathological diagnosis was recorded.

In 3 of the 6 cases of dermoid cyst, there were 2 dermoid cysts, completely separate from each other, attached by separate pedicles, and, in addition, a

normal ovary on the opposite side. In all 3 cases with 2 dermoid cysts, the cysts were situated on the same side, unilateral.

Some of these tumors were large, reaching the xyphoid; others were no larger than walnuts. Some were retroperitoneal, showing no connection whatever with the pelvic organs; others were attached to the uterus or broad ligaments by pedicles of various lengths.

In one instance, reported in 1940, the author (Way) recorded a case of a large tumor which he found in the lower abdomen in a woman upon whom he had done a hysterectomy and bilateral oophorectomy for an ovarian tumor in 1925. Because this second tumor derived its blood supply directly from the ovarian vessels, he considered the possibility that it might have arisen from a third ovary, although no ovarian tissue was found in it. He also stated that it might have been a recurrent tumor, arising from ovarian tissue which he may not have removed at the first operation. This situation has arisen several times, and we mention the cases but do not include them in our series.

Whether a pelvic or abdominal tumor may be presumed to have arisen from an ovary merely because it is of a type that often has that origin is a problem in pathology. Hence, I have referred that question to Dr. Arnold Rich, Emeritus Professor of Pathology at the Johns Hopkins University, and to Dr. Donald Woodruff, in charge of the Laboratory of Gynecological Pathology at the same institution. Independently, they reached the same conclusion—that one cannot conclude that any of the tumors listed above definitely arose from an ovary, unless one can find ovarian tissue in the tumor. This immediately divides these cases into two groups, which we shall list.

1. *Tumors Which Contained Ovarian Tissue.*—These have been reported by Ruppolt, Thumin, Seitz, and Frank. In these instances, the fact that the tumor contained ovarian tissue proves that it arose from an ovary, which in these cases must have been a third ovary. We are therefore adding these to the cases of accessory ovary. Because of the occasional distortion of the pelvic anatomy by the tumor and because of occasional lack of pertinent data, we are not classifying them as supernumerary ovaries.

2. *Tumors Which Did Not Contain Ovarian Tissue.*—These were reported by Charcot, Winkler, Dorn, Lumniczer, Winkel, Baumgarten, Fischel, Olshausen, Sippel, Bassini, Verfasser, Liegner, Vierarm, Schultz-Schultzenstein, Rosenstein, Franz, Stolz, Neumann, Galabin, Furniss, Kriss, and Gardner, Green and Peckham.

In practically every one of the above instances, the author reported his case as one of supernumerary or accessory ovary which gave rise to the tumor. Gardner, Green, and Peckham found their case of granulosa-theca cell tumor, 3.5 cm. in diameter, in the broad ligament in a woman, 64 years old, with carcinoma of the endometrium. This case was included in their study of tumors of the broad ligament, and they assumed that it arose from a bit of ectopic ovarian tissue.

Associated congenital malformations were found in only 2 of these 26 reports. Ruppolt found a bifid Fallopian tube in his case of dermoid cyst which contained ovarian tissue; Neumann found an accessory Fallopian tube in a case of malignant dermoid cyst, which did not contain ovarian tissue. The incidence of associated congenital malformations is, therefore, much lower than in the group of true supernumerary, accessory, and lobulated ovaries.

Undoubtedly, in some of these cases of pelvic tumor in women who had in addition 2 ovaries, the tumor may have arisen from a supernumerary or accessory ovary, even though the tumor contained no ovarian tissue. We bear this possibility in mind in presenting this pathological classification.

The relatively large number of these tumors and the probability that some of them arose definitely from ectopic ovarian tissue suggest the possibility that accessory and supernumerary ovaries may not be as rare as we suppose. It also seems to indicate that the incidence of neoplastic change in such ovaries is much greater than in normal ovaries. The exact incidence would be difficult to determine.

Menstruation and Pregnancy After Bilateral Oophorectomy

The literature contains 4 reports of women who continued to menstruate after bilateral oophorectomy (Schantz, Malcolm, Rouffart, and Manton). It also records 2 instances in which women conceived after bilateral oophorectomy (Hoegh, and Malcolm).

The reports concerning menstruation and pregnancy after bilateral oophorectomy are very sketchy. In spite of this, it is certain that menstruation or uterine bleeding of some sort persisted quite often after such operations, much more often than was reported. It should be noted that "bilateral ovariectomy" was done for a great variety of disorders—myomas, functional bleeding of all sorts, and various general complaints. In many such conditions, the removal of the ovaries would have no effect on the uterine bleeding. In some recorded cases, the women were postmenopausal.

In 1887, John Homans, II, noted the frequency with which bilateral oophorectomy "by various native and foreign operators" had no influence whatever on menstruation. He quoted Goodman, who reported in the *Louisville Medical Journal* in 1875, that following 27 such operations performed by various surgeons, 10 patients continued to menstruate. Homans stated that in more recent reports the record improved, although in 117 more recent cases, 8 patients continued to menstruate. This persistence of menstruation was often attributed to "supplementary ovaries." Homans stated that he did not care to discuss the advisability or indication for Battey's or Tait's operation; he stated that it was being performed for the treatment of a "bleeding fibroid or for the relief of ovarian neuralgia or of mental or moral symptoms." Nor did he suggest the possibility that the persistence of the reproductive and menstrual function after bilateral oophorectomy might be due to the incomplete removal of the ovaries. He attributed it to supernumerary ovaries.

Malcolm, however, in 1903, attributed this persistence of menstruation after bilateral oophorectomy to incomplete operation. He cited 3 cases of his own, in premenopausal women, in which he operated a second time and found ovarian tissue which apparently he had not removed at the first operation. He also reported 2 cases of postmenopausal women in which he operated a second time to remove ovarian tumors which had recurred after bilateral oophorectomy. These recurrences were also accompanied by uterine bleeding. Malcolm failed to distinguish the regular cyclic bleeding in premenopausal women from the pathological irregular bleeding which may accompany any ovarian tumor. Nor did anyone at that time know of granulosa cell tumors or other tumors which secrete hormones and which may produce uterine bleeding.

Consequently, the reports concerning the persistence of uterine function after bilateral oophorectomy are difficult to evaluate. We are inclined to share the opinion of some contemporary surgeons that in most instances it was due to incomplete removal of the ovaries rather than the presence of supplementary or accessory ovaries. As one of the problems that concerned gynecologists before the advent of hysterectomy, and because it was frequently attributed to supernumerary or accessory ovaries, we are mentioning it for its historic interest.

Embryology

The method of origin of these malformations of the ovaries has been discussed in most of the articles on this subject since Grohe's first published case in 1864. In general, on the basis of clinical findings, these cases seem to fall into two groups, supernumerary and accessory ovaries. The group of lobulated ovaries seem to form a class of its own.

As we have indicated, in the rare cases of supernumerary ovary (4 in all), the fact that the supernumerary ovary is completely separate from and not in any way connected with the normally placed ovary has suggested to students of this problem that it has an independent origin embryologically. This seems fairly clear in those cases in which the supernumerary ovary is not located in the pelvis at all or has no connection whatever with the pelvic organs. This explanation is further supported in those cases in which there are associated congenital malformations of other organs that originate in the urogenital ridge.

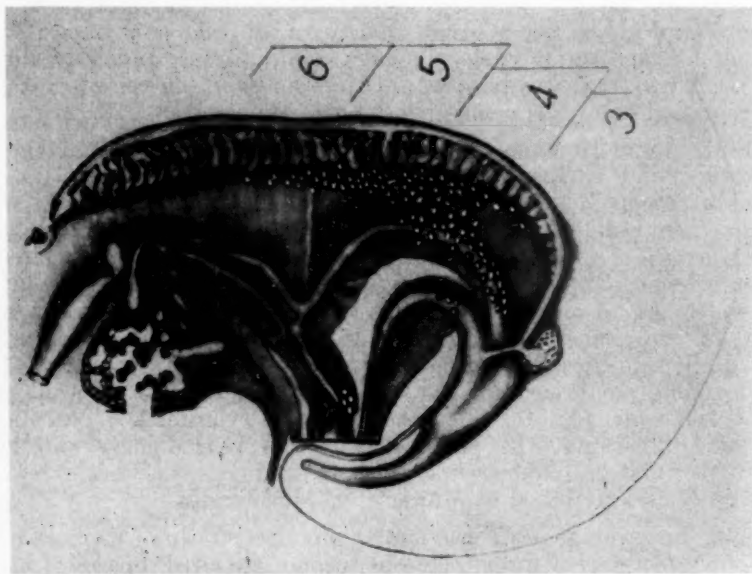


Fig. 9.—The nephric and endodermal organs with their mesenteries of the lower part of a human embryo of 38 somites. The migrating germ cells are shown as dots. The majority are in the mesentery of the hind gut and the celomic angle. Some are still in the endoderm of the intestine. Witschi calls particular attention to two clusters of extraregional gonia, one at the base of the allantoic stalk, the other in the umbilical part of the yolk stalk. (From Witschi. Courtesy of Contributions to Embryology, Carnegie Institute of Washington.)

A second possible explanation of the development of supernumerary ovaries is suggested by the work of Witschi on the migration of germ cells in the human embryo. In describing the earliest life history of the germ cells, Witschi found that "in the youngest embryo all germ cells are in the endoderm of the yolk sac near the allantoic evagination. . . . In embryos of from 13 to 20 somites, the germ cells become transferred from the yolk sac to the hind gut. A variable number—probably always less than half—remain temporarily in the endodermal epithelium. The larger number leave the endoderm and move into the mesenchyme. . . . The second phase of migration begins in embryos of more than 25 somites. The germ cells leave the gut, ascend through the mesentery, and advance laterally toward the mesonephric folds. . . . Germ cells reach their positions in the cortex of the primitive gonads

by different routes. . . . During the entire migratory period the number of germ cells increases steadily through mitotic divisions." As has been pointed out, the greatest distance that any germ cell traverses in this migration is not more than 0.5 mm., because of the shortness of the mesentery of the hind gut, the proximity of the structures, and the small size of the embryo. The illustration taken from Witschi's article (Fig. 9) is a representation of the nephric and endodermal organs, with their mesenteries, of the lower end of the embryo. The migrating germ cells are shown, their size somewhat exaggerated. Two clusters of extraregional gonidia are present in this embryo—one at the base of the allantoic stalk and the other in the umbilical part of the yolk sac.

To the student of supernumerary ovaries, the work of Witschi and this illustration are particularly suggestive for several reasons. In the first place, the germ cells are normally found in the early embryo in precisely the same situations where supernumerary ovaries have been discovered in later life. In the second place, there is a very wide distribution in the normal location of these germ cells. Third, Witschi found that there was occasionally a delay or failure to migrate on the part of some of these germ cells. Fourth, Witschi noted that, whereas some of the extraregional gonidia that had missed migrating showed signs of degenerating, others were nearly normal.

These embryological observations suggest a possible explanation for the formation of supernumerary and possibly accessory ovaries. Supernumerary ovaries have been found in the same situations in which the germ cells spent part of their early life, and from which they normally migrated. If any of these germ cells failed to migrate and continued to live and multiply in the mesentery of the sigmoid or retroperitoneal areas and if the surrounding mesenchyme became transformed into ovarian stroma, the result would be a supernumerary ovary.

This explanation, however, involves several assumptions which have not been proved. Embryologists do not know whether germ cells can live, multiply, and acquire maturity outside the nutritive environment of the primitive gonad. Also, they have pointed out that there is no evidence, except the 2 cases of supernumerary ovary we have presented, that the mesenchyme around the germ cells could be transformed into ovarian stroma.

In spite of the lack of supporting evidence, however, they suggested that this possible explanation of the origin of supernumerary ovaries is worth consideration.

As Burns has indicated, supernumerary ovary is such a rare condition that not only clinicians but also embryologists have been unaware of its existence. Hence, it has received neither clinical nor embryological investigation. It is almost a certainty that more cases will be discovered and that the clinical problems suggested will stimulate embryological investigation.

The origin of accessory ovaries has been attributed to a simpler procedure, the separation of bits of tissue from the primordium of the ovary or from the developing ovary. From the clinical evidence, it does not seem necessary to postulate a separate primordium, as seems to be the case with the true supernumerary ovary.

One of the chief problems that concerned the older students of this subject was the determination of the factors that could cause bits of the embryonic ovary to break off and pursue an independent existence. They listed among these factors the following: (1) fetal peritonitis, producing adhesions which might divide the embryonic ovary; (2) torsion of the pedicle of the embryonic ovary, thus interfering with its blood supply and possibly separating parts of the ovary; (3) pressure of surrounding organs; (4) pelvic tumors; and (5) syphilis.

Hence, in almost all detailed studies of this subject in past years, careful examinations were made to discover any of these factors, especially pelvic adhesions or any evidence of fetal peritonitis. In more recent articles, these factors have lost their significance.

Supernumerary Ovaries in Lower Species

In assembling the references to supernumerary and accessory ovaries in human beings, the staff of the Welch Library also noted all references to similar conditions found in animals and lower species. We present this list for its biological interest.

In 1951, Krohn reported a case of endometriosis and supernumerary ovary in a rhesus monkey. In 1932, Blotevogel reported a supernumerary ovary in an ape, and in 1946, Culiner found one in a baboon. Shaw and Brambell reported an aberrant ovary in a frog, and in 1924, de Luna presented before the French Academy of Sciences a report of an accessory ovary found in the fruit fly. These unusual findings are of interest to gynecologists as well as to biologists.

Summary

Two cases of supernumerary ovary and one of accessory ovary, proved by microscopic examination, have been reported.

Rokitansky in 1861, and Klob in 1864, stated that no case of supernumerary ovary had ever been reported. The first case of accessory ovary was reported by Grohe in 1864.

A review of the medical literature since 1861 reveals only 2 cases of supernumerary ovary and 18 reports of accessory ovary. The distinction between the 2 is at times uncertain and there have been differences of opinion concerning classification.

Supernumerary ovary is one of the rarest of gynecological conditions, its exact incidence being difficult to estimate. Only one case has been found in 29,000 autopsies and among all the clinical records of the Johns Hopkins Hospital. There has been only one case in over 400,000 admissions to the Hospital for the Women of Maryland.

Accessory ovary is also quite rare; there has been only one recognized case in 93,000 gynecological admissions.

The congenital malformations associated with these anomalies have been listed. Other conditions attributed to supernumerary or accessory ovaries, such as pelvic tumors and persistence of menstrual and reproductive functions after bilateral "ovariotomy," have been presented and discussed.

The reported cases of supernumerary ovary found in lower animals have been listed.

A brief discussion of the embryology of this condition has also been presented.

I wish to acknowledge my indebtedness to the following persons: the staff of the Welch Library; the staff of the Department of Embryology of the Carnegie Institute of Washington; Dr. Arnold Rich, Emeritus Professor of Pathology; Dr. Donald Woodruff, in charge of the Laboratory of Gynecological Pathology; and Dr. Richard W. Te Linde, Professor of Gynecology.

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Discussion

DR. ANDREW A. MARCHETTI, Washington, D. C.—It appears to me that as rarely as the anomaly of supernumerary ovary has been authenticated, Dr. Wharton is now in the unique position of having encountered it on 2 occasions in recent years and in both cases was able to establish its authenticity.

I should like to make one brief comment upon the classification of supernumerary and accessory ovaries as it was considered in the paper. The author pointed out that at times the differentiation between a supernumerary ovary and an accessory ovary is not easy. However, he does mention that in the clearest cases of supernumerary ovaries reported in the past, the anomaly was found to develop in retroperitoneal regions. I wonder if it would not enhance the classification if the retroperitoneal position were added to the other basic prerequisites which were enumerated for establishing the existence of a supernumerary ovary.

Dr. Wharton's paper leaves me with the thought that accessory ovarian tissue could be found more often if we searched for it; but not the supernumerary ovary.

DR. ARTHUR HERTIG, Boston, Mass.—There were two interesting things to me in this fascinating paper: (1) the general location of these accessory bits of ovarian tissue, and (2) the concept that perhaps the migrating germ cell induced these bits of ovarian tissue in their ectopic location.

I think one should point out that most of the people who have worked on the problem of the migrating germ cell in later years, including our own group, agree that there are migrating germ cells. We believe that they are set aside or are first seen in the endodermal region very early in the life of the segmented egg.

If one carries this concept along, one realizes that the presence of teratomas throughout the retroperitoneal midline region up to the pineal can apparently be explained or is consistent with the explanation that these aberrant germ cells do give rise to teratomas of one or more elements. Most of those do not seem to have any accessory ovarian stroma associated with them. My own feeling would be that a misplaced germ cell might give rise to a retroperitoneal teratoma, but that accessory ovarian tissue can only occur in the general region of the urogenital ridge where gonadal stroma is formed.

DR. WHARTON (Closing).—There was one item I did not mention and that was the incidence of supernumerary ovaries. We have had 2. One was from the old Woman's Hospital and was the only case of its kind there; the case number was over 400,000. The second was from Johns Hopkins and the case number was over 700,000. Of course this is not an accurate way of estimating the incidence, and the incidence at autopsy is more significant. Dr. Rich found only 1 in 29,000 autopsies, however, which indicates that a thorough study also does not often turn them up.

I appreciate Dr. Marchetti's suggestion. I wonder if he knows how many man hours I will have to spend now because of that suggestion because I will have to go over the records I have and see how many had retroperitoneal masses.

I appreciate Dr. Hertig's suggestion of the origin of the supernumerary ovary. The location is exactly in accordance with anatomy and embryology.



Gynecology

THE MOVEMENT OF TRITIUM-LABELED WATER IN NEOPLASTIC CYSTIC STRUCTURES*

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NEOPLASTIC ovarian cysts can usually be distinguished from their non-neoplastic counterparts by the presence of a distinct fibrous wall which grows with the tumor. There is also a lining epithelium which varies with the type of tumor and in the case of multilocular tumors may vary from locule to locule. To many of us these morphologic characteristics and the extremely slow growth or even static nature of some of these cysts have suggested loculation or segregation of the cyst contents from the general body fluids. Further, the variable nature of their fluid contents both physically and chemically implies active secretion, generally a one-way process, as the most likely mechanism for production of the contained fluid. Such loculation or static separation from the vascular compartment has not been found true of nonneoplastic cysts^{1, 2} and other body fluids.^{3, 4} For this reason it seemed of interest to determine the permeability to tritium-labeled water of the barrier between the blood and cyst fluid.

Material and Methods

Eight women with neoplastic cysts of various types and sizes have been studied. In addition, one patient with hydrosalpinx and one with two small sessile, very thin-walled broad ligament cysts were studied. Between 1.5 and 5 mc. of tritiated water was administered intravenously at variable times prior to laparotomy. Heparinized venous blood samples were obtained at appropriate intervals during the course of the experiment. The plasma was separated from these samples and retained for counting. Experimentation on excised specimens demonstrated that a 20 gauge needle could be passed obliquely

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through the wall of such cystic structures without evident leakage of fluid. This method was used in situ to obtain tumor fluid for counting after the tumor was isolated as much as possible from the operative field. Samples ranging from 2 to 10 ml. were removed at intervals of approximately 10 minutes from one or more locules prior to excision of the tumor, the number of consecutive samples being limited by the time available. A few grossly bloody samples were discarded and the remainder were counted in a Packard Tri-Carb Model 314-DC Liquid Scintillation Spectrometer, the background varying from 68 to 88 disintegrations per minute (DPM).²

Results

The gross and microscopic characteristics of the cystic structures studied are recorded in Table I. Four of these tumors were benign serous cystomas

TABLE I. CHARACTER OF CYSTIC STRUCTURES

PATIENT	TYPE OF TUMOR	EXTERNAL SIZE (CM.)	EPITHELIAL LINING	FLUID	
				VOLUME (ML.)	APPEARANCE
D. B. W.	Cystoma	10 × 8 × 6	Serous	253	Clear, watery
E. M.	Cystoma	10 × 8 × 7	Serous	279	Clear, watery
D. B.	Cystoma	12 × 8 × 8	Serous	400*	Clear, watery
H. S.	Cystoma	5	Serous	58	Clear, watery
H. C.	Hydrosalpinx	16 × 6 × 6	Serous	300*	Clear, watery
M. S.	Cystadenoma	19 × 29 × 11	No. 1, pseudomucinous	180	Mucinous yellow turbid
			No. 2, pseudomucinous	2,140	Serous white turbid
			No. 3, pseudomucinous	320	Serous white turbid
I. A.	Cystadenocarcinoma highly loculated	30 × 20 × 20	No. 1, pseudomucinous	30*	Highly mucinous white turbid
			No. 2, pseudomucinous	30*	More mucoid—micropipetting impossible
V. E.	Dermoid	8 × 7 × 6	Stratified columnar with stratified squamous	170	Clear, watery
B. P.	Dermoid	6 × 5 × 5	Stratified squamous with sebaceous glands and hair	80	Sebaceous—few hairs
M. H.	Broad ligament cysts	2 × 1.0	Not determined	3*	Clear, watery
		2 × 0.5		2*	Clear, watery

*Estimated from measurements.

(unilocular) and were lined by the various types of epithelia usually seen in such cysts (Figs. 1 and 2). There were 2 pseudomucinous tumors (Figs. 3 and 4). Both were cystadenomas (multilocular), one appeared benign, and the other microscopically malignant. There were 4 large locules in the benign tumor (Patient M. S.), one of which showed evidence of previous tapping (a paracentesis had been attempted elsewhere). For this reason only 3 of the locules were studied. The malignant pseudomucinous tumor contained a large number of small locules, only 2 of which were sampled. The fluid from one was so viscous that it could not be accurately pipetted, though the tritium concentrations in both locules were within the same general range.

There were 2 cystic teratomas or dermoids. Both were multilocular, one locule being much larger than the rest in each instance. In Patient B. P. this

cavity was lined by the usual stratified squamous epithelium with sebaceous glands, occasional sweat glands, and hair. The contents of this cyst were primarily sebaceous. In the other the cavity was lined by a multilayered epithelium of stratified columnar appearance (Fig. 5) in some areas and stratified squamous epithelium in others. The former epithelium appeared metaplastic in

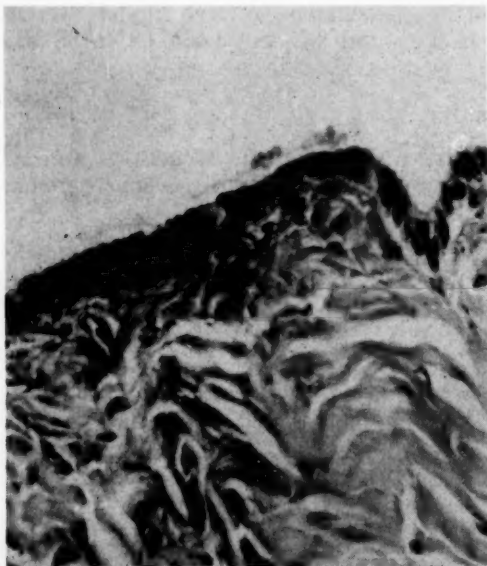


Fig. 1.



Fig. 2.

Fig. 1.—Lining of the serous cystoma from Patient H. S.

Fig. 2.—Lining of the serous cystoma from Patient D. B.

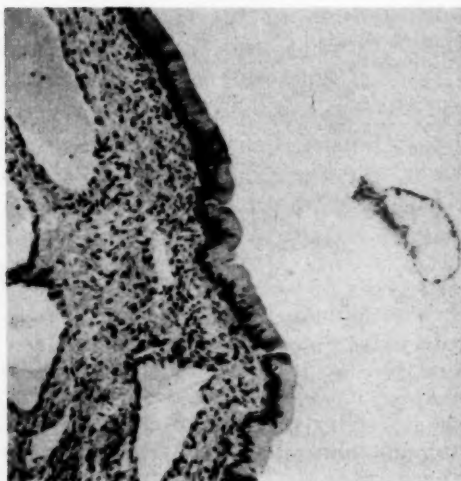


Fig. 3.

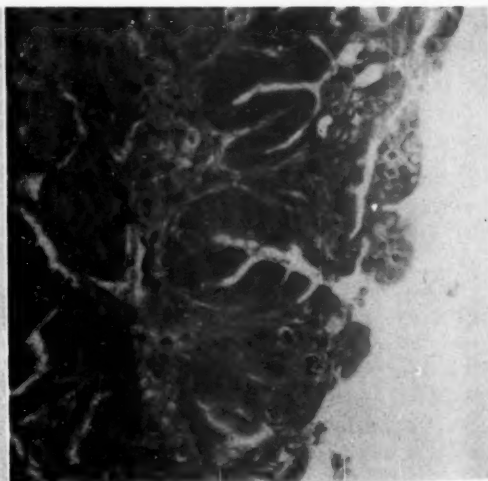


Fig. 4.

Fig. 3.—Lining of the pseudomucinous cystadenoma from Patient M. S.

Fig. 4.—Lining of the pseudomucinous cystadenocarcinoma from Patient I. A. This epithelium varied from obviously benign in some areas to grossly malignant in others.

some places. The fluid from this cyst was clear and watery. The hydrosalpinx was not unusual in appearance; the exact histologic nature of the small broad ligament cysts is uncertain because of the trauma of aspiration. Grossly, they were quite vascular and there were no peritoneal adhesions in the pelvis.

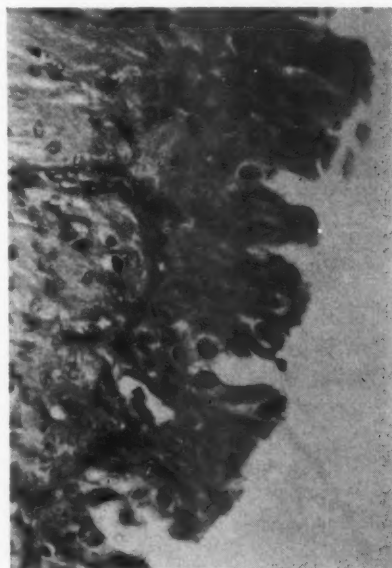


Fig. 5.—Lining of the dermoid from Patient V. E. In other areas this cyst was lined by stratified squamous epithelium.

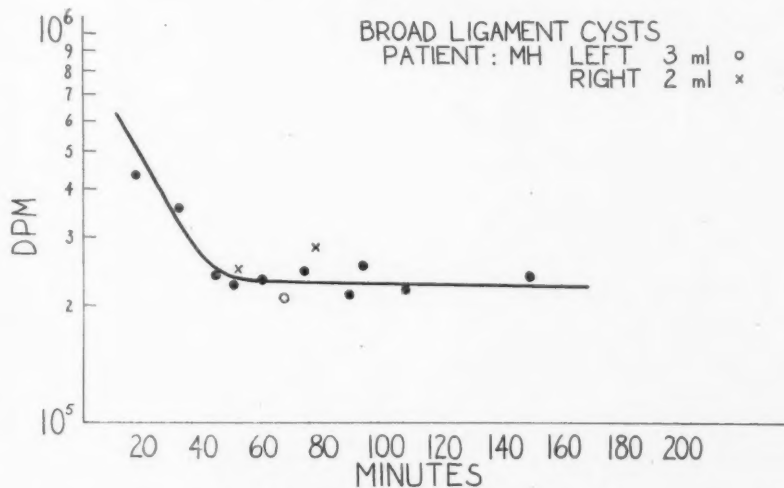


Fig. 6.—Patient M. H. Peripheral venous dilution curve and cyst fluid concentrations for tritiated water after intravenous injection (in disintegrations per minute). The cyst samples, which have equilibrated with the blood, are single samples.

It is apparent from the venous dilution curve in Fig. 6 (Patient M. H.) that the intravenously administered tritiated water equilibrated with body fluids in approximately 45 minutes, about one hour being average for the entire group

of patients. The shape of this curve during the first 40 to 75 minutes varies from patient to patient, probably because of variable tissue equilibration rates.⁵ Plotted on this curve are the tritium concentrations found in the fluid from

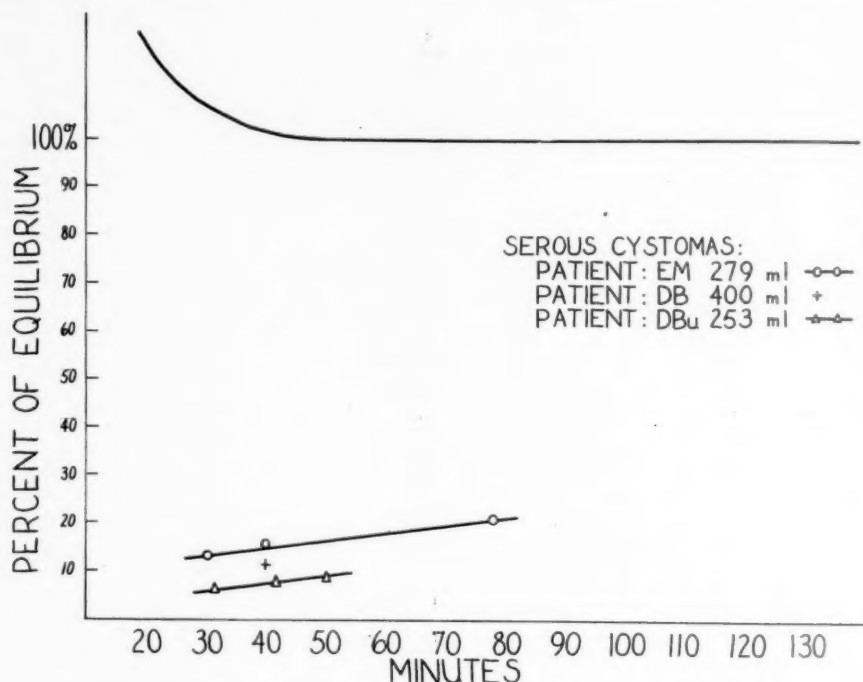


Fig. 7.—Concentrations of tritiated water in the cyst fluids in the first 2 hours plotted as per cent of blood equilibration value for each patient. The dilution curve is purely empiric.

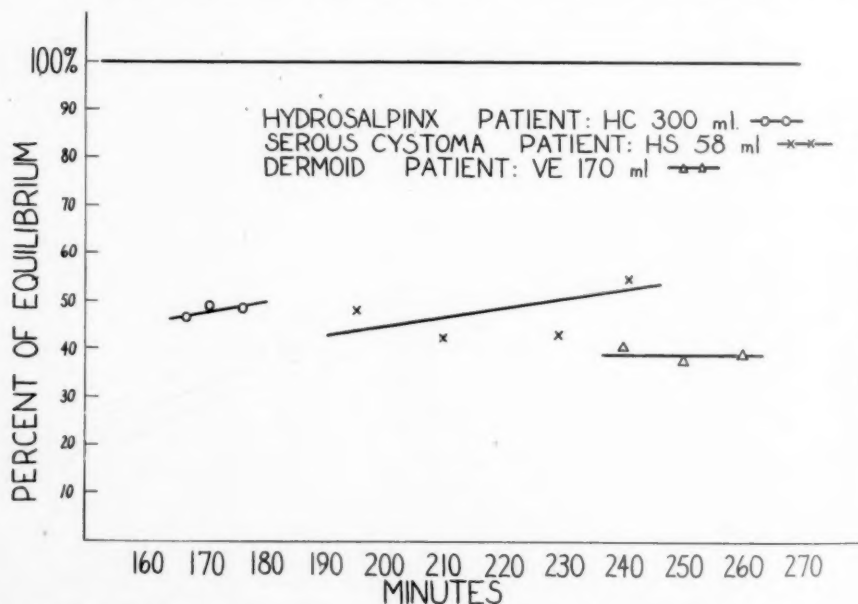


Fig. 8.—Concentrations of tritiated water in miscellaneous cystic structures, all with clear, watery fluid.

the very small broad ligament cysts. These fluids had obviously equilibrated with the blood 54 minutes after tracer administration, suggesting an exchange rate in the same range as that seen in the ovarian follicle.

Fig. 7 portrays the data from the 3 serous cystomas sampled within the first 2 hours after administration of the tracer. These data have been plotted as the per cent of the blood equilibration value for each patient, the venous dilution curve being purely an empiric. Again the exchange rate is demonstrated to be extremely rapid, as significant amounts of tritium were present in all 3 of these tumors. Despite the variations in fluid volume the concentrations were all in the same general range.

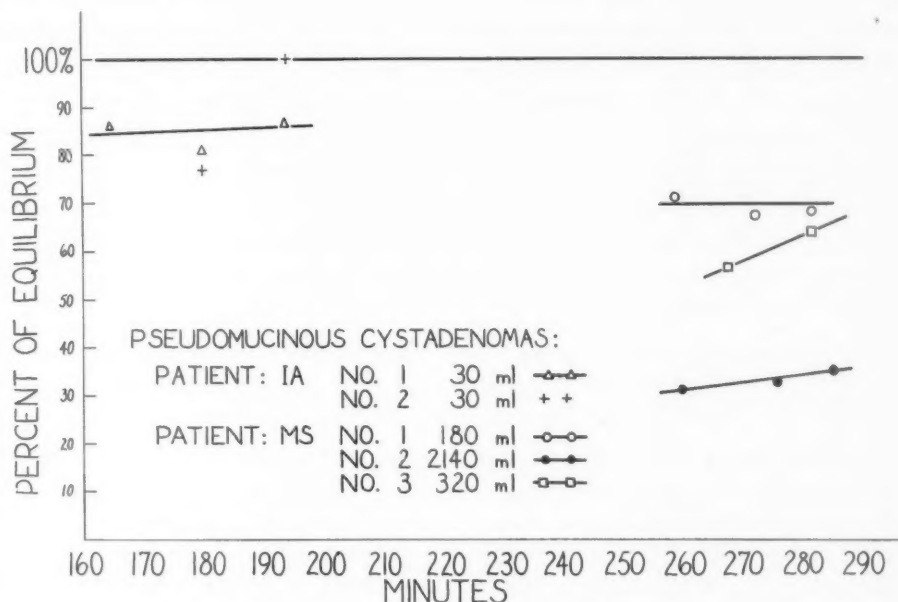


Fig. 9.—Concentrations of tritiated water in cyst fluid from various locules in two multilocular pseudomucinous tumors. The wide variation in the two samples from locule No. 1 of Patient I. A. is due to pipetting difficulties (see text).

The data for the remaining cystic structures have been plotted in a similar manner (Figs. 8 and 9). These samples of cyst fluid were all obtained in the third, fourth, and fifth hours after administration of the tracer. Significant amounts of tritium were found in all cysts with fluid contents. The sebaceous material from one of the dermoids (Patient B. P.), which was solid at room temperature, did not contain demonstrable tritium. In the multilocular cyst from Patient M. S., it was possible to determine the tritium concentration simultaneously for each locule. As may be seen in Fig. 9, the concentrations varied inversely with the total volume of the fluid contents. It seems likely that the lower concentration of tritium in the largest locule (No. 2) did not result from any difference in "membrane" permeability but resulted from the prolonged mixing time due to the larger volume of this locule.

Comment

It seems clear that the movement of tritiated water from the vascular space into the cyst contents is very rapid regardless of the character of the cyst lining, provided the contents contain a high proportion of water. Since the

cyst volume does not change materially during the period of observation, this movement must be a matter of exchange. The mechanism most likely involved is simple diffusion, the barrier between the cyst fluid and the vascular compartment being highly permeable to water molecules. These data do not give information concerning fluid accumulation or *net* transfer of fluid into such cystic cavities. It is apparent, however, that the epithelial lining and connective tissue wall of these cysts do not constitute an impervious barrier to the movement of fluids. While secretion from lining cells certainly contributes to their fluid contents, judging from the variable character of the solutes in such fluids,⁶ it may well be that the primary mechanism of fluid collection is on the basis of changes in hydrostatic and osmotic pressures, particularly where rapid changes are seen.

Summary and Conclusions

The permeability to water of the barrier between the blood and cyst fluids of various cystic structures has been investigated using tritiated water as a tracer. Among the 10 patients there were 4 with serous cystomas, 2 with pseudomucinous cystadenomas, 2 with cystic teratomas (dermoids), one with broad ligament cysts, and one with a hydrosalpinx. Tritiated water appeared in the cyst contents promptly in all but one.

All samples were counted by Mrs. Lynn Gilboe under the direction of Dr. Charles Heidelberger in the McArdle Cancer Research Laboratories. We are most grateful to Dr. Heidelberger and his staff for making these studies possible.

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CARCINOMA OF THE CORPUS UTERI: DIAGNOSIS AND THERAPY*

H. L. KOTTMEIER, M.D., STOCKHOLM, SWEDEN

(From the Radiumhemmet)

CARCINOMA arising in the endometrium is the second most frequent malignant tumor to develop within the genital organs of women. There is mounting evidence that there is an absolute increase in the incidence of neoplasm of the corpus uteri. This fact can mainly be attributed to the marked increase in life expectancy. Carcinoma of the corpus, in particular, is a disease of postmenopausal age and appears exceptionally in women under 40 years of age. While, in Sweden, the ratio of fundal to cervical malignancies is 1:6 in patients prior to menopause, it is approaching 3:1 in patients who are in their seventies.

An investigation in Stockholm of the cause of postmenopausal vaginal bleeding indicates that 6 out of 10 patients who consult a doctor because of spontaneous bleeding 2 years or more after the menopause have a malignant tumor of the pelvis. Most patients with endometrial carcinoma are suffering from bleeding or bloody discharge. In the years 1948 through 1951, we have seen 617 patients with primary carcinoma of the endometrium at the Radiumhemmet. Menstruation had ceased in 508 of the patients, in 41 per cent of them at the age of 51 or more.

In spite of these facts, the delay in diagnosis of the carcinoma is formidable. Seventy per cent of the patients had had symptoms for more than 3 months and 23 per cent for more than one year. Delay due to the physician was verified in more than 10 per cent of the cases. In several patients no pelvic examination had been carried out. Others had been treated for a senile vaginitis or cervical polyps, easily removed. We agree with French authorities that a cervical polyp at postmenopausal age is often a sign of an endometrial carcinoma. In the propaganda for early diagnosis the teaching of physicians is still important.

Pathogenesis

Carcinoma of the corpus is often accompanied by degenerative cardiovascular disease, essential hypertension, obesity, diabetes, and/or other endocrine abnormalities. Of the Radiumhemmet's 617 patients, 7 per cent weighed more than 220 pounds, 36 per cent more than 180 pounds. Twenty-three per cent of the patients were unable to perform any physical activities without discomfort because of cardiovascular disease. Seven per cent had diabetes. These and other facts justify the suggestion that endocrine factors play a part in the pathogenesis of endometrial carcinoma.

*Presented as Joseph L. Baer Lecture before the Chicago Gynecological Society, Oct. 18, 1958.

There is evidence that prolonged estrogen stimulation of the endometrium occurs frequently in women who develop adenocarcinoma of the corpus. For instance, the average age of menopause is 51.6 in our cases of corpus carcinoma while in general it is 48.7 among Swedish women.

Many clinicians state that there is an association between cancer of the corpus and endometrial hyperplasia, and that quite a few patients with adenocarcinoma of the endometrium previously have suffered from dysfunctional uterine hemorrhage. This has not been our experience. As a matter of fact, 104 patients suffering from recurrent functional uterine hemorrhage were

Fig. 1.

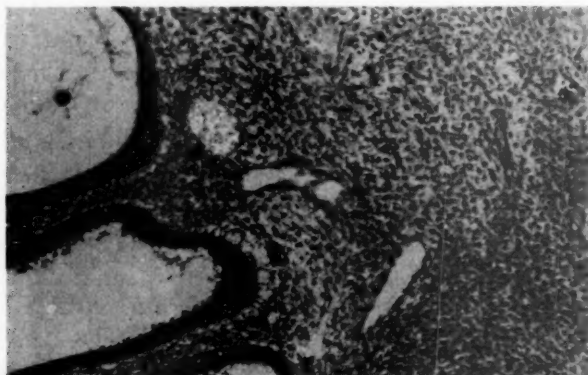


Fig. 2.

Fig. 1.—Postmenopausal endometrial hyperplasia of Swiss-cheese type. Sign of marked estrogen stimulation. ($\times 160$; reduced $\frac{1}{5}$.)

Fig. 2.—Postmenopausal endometrial hyperplasia with a dense and firm stroma and thick-walled vessels. ($\times 160$; reduced $\frac{1}{5}$.)

referred for radiation sterilization to the Radiumhemmet between 1918 and 1947 but did not receive any irradiation. A carcinoma of the corpus was diagnosed later in 4 of these 104 cases, i.e., in 3.9 per cent.

Of considerable interest is the observation by Hertig¹³ that an endometrium characterized by few or many endometrial glands composed of large cells with abundant clear eosinophilic cytoplasm is precancerous in nature. We share this opinion and allot these cases to Group IX in the Radiumhemmet classification.

Several authors have stated that there is a developmental relationship between an endometrial hyperplasia persisting into the postmenopausal age group and endometrial carcinoma. To some extent, our experience differs

from this point of view. To understand the pathogenesis of postmenopausal endometrial hyperplasia it is important, in our opinion, to pay attention to the endometrial vessels and the stroma by the use of special staining. Cases of endometrial hyperplasia, sometimes the Swiss-cheese type with hyperproliferation of the epithelium, the glands, and the stroma, should be differentiated from cases of endometrial hyperplasia with an often dense and firm stroma frequently rich in collagen fibrils and with thick-walled endometrial vessels suggesting poor estrogen activity (Figs. 1 and 2). This endometrial pattern may be regarded as hyperplastic polyps or an area of localized hyperplasia, in which case the pathogenesis is obviously different from that of a typical Swiss-cheese endometrium. An ovarian tumor, usually a granulosa cell or theca cell tumor, has been demonstrated in 92.9 per cent of 148 patients with typical Swiss-cheese hyperplasia in whom menstruation has ceased for 2 years or more. This type of endometrial hyperplasia could be demonstrated in the curettings from 5 of 508 postmenopausal patients with carcinoma of the corpus. In 2 of these 5 patients a theca cell tumor was diagnosed. A postmenopausal endometrial hyperplasia with dense and firm stroma was diagnosed in 91 of the 508 patients mentioned. An ovarian tumor has not been diagnosed in any of the 91 cases. These observations prove the importance of distinguishing between different types of postmenstrual endometrial hyperplasia.

Smith⁴³ has pointed out that a hyperplasia of the connective tissue of the ovarian cortex remarkably often is associated with an adenocarcinoma of the endometrium. McKay³⁰ reports cortical stromal hyperplasia in 88 per cent of 331 cases of corpus carcinoma and a similar cortical hyperplasia in 44 per cent of 307 control cases. The Radiumhemmet series of cases of corpus cancer is not appropriate to such an investigation since most of the cases received radiotherapy prior to operation. Generally, we have not observed that cortical stromal hyperplasia is more common among the patients with corpus cancer than among other patients of postmenopausal age.

The reports on adenocarcinoma of the endometrium in association with the Stein-Leventhal syndrome and with granulosa cell tumors and thecoma also speak in favor of hormonal factors playing a part in the pathogenesis of corpus cancer. Speert⁴⁴ reports that 14.8 per cent of cases with feminizing ovarian tumors are associated with endometrial carcinoma. Hertig states that about 20 per cent of the patients with granulosa cell or theca cell tumors of the ovary have associated carcinoma of the corpus. We agree with Robert Meyer, McKelvey,³¹ and others that the association of adenocarcinoma of the endometrium and estrogen-producing ovarian tumors is not so common as often pointed out, even if we do not share the opinion of Emge⁹ that the association is not more frequent than could be explained by chance alone. An adenocarcinoma occurred in 3 out of 99 granulosa cell tumors and in 4 out of 26 theca cell tumors seen at the Radiumhemmet through the year 1951. Moreover, endometrial atypicalities, suspected to be of carcinomatous nature, have been diagnosed in 11 other of the 125 cases, but the endometrial pattern could not be referred to as obvious adenocarcinoma in our opinion.

Pathologic Considerations

Primary carcinomas of the corpus are generally adenocarcinomas. A pure solid carcinoma was diagnosed in 2.5 per cent and squamous epithelial carcinoma in 0.4 per cent of the Radiumhemmet series. Mixed tumors are rare.

Most so-called carcinosarcomas occurring in the endometrium are, we believe, anaplastic adenocarcinomas and not real mixed tumors. Sometimes it is difficult to distinguish a carcinoma from a myosarcoma.

Adenocarcinomas of the endometrium may vary in degree of differentiation. Several authors have pointed out that a poorly differentiated tumor has

a greater tendency to destroy deeply the myometrium than a highly differentiated one. Lindgren²⁴ has confirmed this observation in a study of 525 patients with carcinoma of the endometrium operated on at various hospitals and later referred to the Radiumhemmet for irradiation. The ratio of superficially infiltrating carcinoma to deeply infiltrating carcinoma was 3:1 in the highly differentiated adenocarcinomas while in the more anaplastic adenocarcinomas the ratio was 1.5:1. The difference is statistically significant. The careful studies by Long and Taylor²⁶ on the relation of the size and multiplicity of the nucleoli to the degree of differentiation of the adenocarcinoma and the prognosis are of considerable value. I believe these facts are of interest relating

Fig. 3.

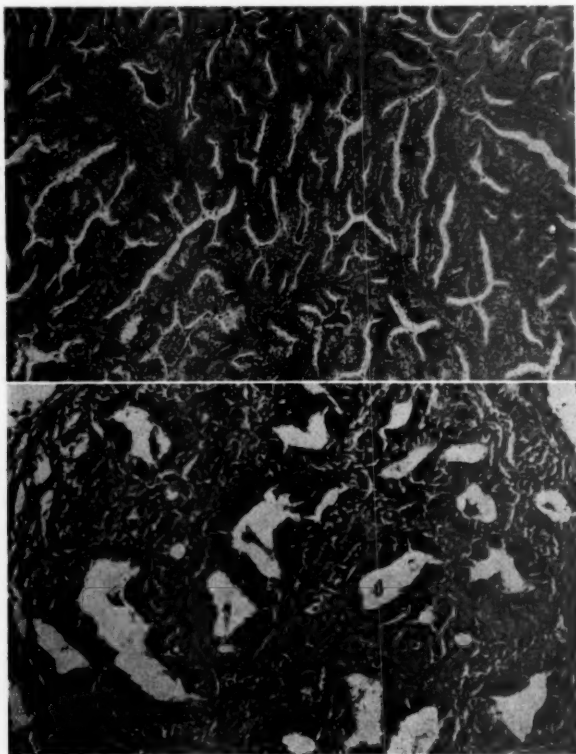


Fig. 4.

Fig. 3.—Curettage. Extremely hyperplastic endometrium suspicious of adenocarcinoma. Patient 33 years of age. ($\times 160$; reduced $\frac{1}{3}$.)

Fig. 4.—Curettage. Atypical endometrial hyperplasia suspicious of adenocarcinoma. ($\times 160$; reduced $\frac{1}{3}$.)

to classification, prognosis, and treatment of carcinoma of the corpus. Statistics on treatment results are of small value if the series of cases are not comparable. Uniformity in statistics on carcinoma of the corpus requires precise definitions of the verities of the disease.

Diagnosis

In general, a curettage is required to establish the diagnosis. An endometrial biopsy is of value in the early diagnosis as it can be performed on an ambulatory basis. The diagnosis of an adenocarcinoma from curettings is

sometimes difficult. The endometrial hyperplasia can on occasions be so violently hyperplastic that it resembles a highly differentiated adenocarcinoma. This holds true particularly in patients about 50 years of age, in patients with ovarian tumors, in patients who have received estrogens in large doses, and in patients of young age. Seventeen of our 1,439 patients with cancer of the corpus (1.2 per cent) were 40 years of age or younger. We believe that some clinicians have exaggerated the occurrence of fundal neoplasms in young women because of misinterpretation of violent endometrial hyperplasia (Figs. 3 and 4). The differential diagnostic difficulties are well demonstrated in 5 patients, 27 to 33 years of age, recently seen at the Radiumhemmet. The curettings from these patients had been interpreted as adenocarcinomatous by pathologists. Four of the patients consulted a physician for irregular bleeding, one for sterility. The diagnosis of an adenocarcinoma was confirmed in one of the 5 cases. The other 4 patients are at present symptom free. One of them has given birth to 2 healthy children.



Fig. 5.

Fig. 5.—Hystero-graphy. Carcinoma of the corpus.



Fig. 6.

Fig. 6.—Hystero-graphy. Carcinoma of the corpus with extension of the growth to the isthmus and ovarian tumor.

The establishment of the diagnosis of a carcinoma by histologic examination of the curettings is unsatisfactory. Prior to the administration of any kind of therapy, we must get information on the extent of the carcinoma to the isthmus and the cervix. In most instances a fractional curettage facilitates the estimation of the extension of the carcinoma. A fractional curettage should be performed in the following order: (1) curettage of the area around the external os; (2) curettage of the endocervical canal; (3) measurement of the length of the uterine cavity; (4) careful dilatation of the cervical canal; (5) curettage of the isthmus; (6) examination of the fundus of the uterine cavity with a cannula forceps; (7) curettage of the fundus and the corpus.

The specimens removed from the different parts of the cavity have to be kept separate and each of them examined microscopically. If the pieces obtained are small, or if it is impossible to be sure clinically that the pieces of tissue were taken from the wall of the endocervix, the correct diagnosis must

depend on whether or not the pathologist is able to prove the presence of both carcinoma and normal cervical glands in the same specimen. This also holds true for the corpus in cases of clinically endocervical carcinoma.

Sometimes the clinical examination or the differential curettage does not give sufficient information regarding the location of the carcinoma. If so,

Fig. 7.



Fig. 8.

Fig. 7.—Hystero-gram. A small cancer in the right uterine corner in a nullipara 30 years of age.

Fig. 8.—Parametro-gram. A well-demarcated tumor in the left parametrium.

hysteroscopy or a hysteroradiographic examination with water-soluble contrast medium is imperative prior to any kind of therapy (Figs. 5-7). Some French and Swedish authorities have stressed the value of the x-ray examination

prior to curettage in every instance of postmenopausal hemorrhage and in every instance of a carcinoma of the endometrium. In excellent studies Norman^{34a} has demonstrated the value of this examination. No doubt the hystero-graphy should be performed in some cases, but at the Radiumhemmet we do not like to do it routinely because we believe it may increase the risk of infection and spread of the cancer. Recently we performed a hysterectomy in a case of a small adenocarcinoma of the corpus one week after the x-ray examination. Metastasis was found in the right ovary straight to the fimbriated end of the tube. The observation is of interest even if it does not prove a direct relation of the metastasis to the x-ray examination. We do not know if free cancer cells in the peritoneal cavity are viable and capable of causing implantation on the peritoneum. Dahle's⁷ findings of tumor cells in wash smears from the peritoneal cavity in 7 out of 21 patients with operable corpus carcinoma are of interest.



Fig. 9.—Venography. Compression of the left external iliac vein by a lymph node.

Special x-ray studies, such as arteriography or venography, or even parametriography by injection of the contrast medium into the parametrium from the vagina, are of value in selected cases of carcinoma of the endometrium associated with another growth in the pelvis. This is demonstrated by a case of extensive adenocarcinoma of the corpus and endocervix with metastases to the vagina. A mass the size of a large plum was felt to the left from the uterus. Roentgen examination verified the suggestion that the tumor was an intraligamentary fibroid (Fig. 8). Venography, however, also revealed compression of the external iliac vein (Fig. 9). Following intensive irradiation by radium

and x-ray for several months, a modified radical hysterectomy and lymphadenectomy were carried out. The mass to the left of the uterus was an intraligamentary fibroid. The compression of the vein was due to lymph node metastasis. Microscopic examination of the lymph node showed pronounced changes in the adenocarcinoma due to the irradiation. There was no cancer remaining in the uterus.

Classification

The present international clinical classification recommends a division of carcinoma of the endometrium into carcinoma of the corpus and carcinoma of the corpus and endocervix. In the Radiumhemmet series this ratio is 4.8:1. A subgrouping of the carcinoma into 2 stages with regard to clinical findings prior to treatment is recommended. A case should be allotted to Stage I if the growth is clinically confined to the uterus, and to Stage II if the cancer has spread outside the uterus. At the Radiumhemmet, we assign a case to Stage II, i.e., inoperable cases, only if the cancer has obviously invaded the surrounding organs or the peritoneum, or if metastases are present. Fixation of the uterus does not permit a case to be allotted to Stage II.

Because carcinoma of the corpus frequently occurs in patients with severe cardiovascular disease or obesity, Heyman¹⁴ recommended the subdividing of the cases classified as Stage I in two groups: Group 1, clinically operable cases; and Group 2, clinically inoperable cases. I agree with Bastiaanse³ that such a staging is not scientific since it depends on the skill and courage of the surgeon. On the other hand, some institutions may receive mainly cases of good operative risk, others of bad risk. In particular this latter fact is valid for radiotherapeutic institutions, such as the Radiumhemmet, for instance, and it will explain the many clinically inoperable cases seen in the years 1948 through 1951 (33 per cent). Only if the anesthetist or the physician considers the patient a really poor risk, have we, since 1948, allotted a case to Group 2. The Radiumhemmet, which is responsible for the radiation therapy of uterine carcinoma diagnosed within two-thirds of Sweden, does not receive for primary therapy all cases of carcinoma of the corpus, since many patients are operated upon at various other hospitals. Our series of cases of carcinoma of the endometrium presented is selected, inasmuch as many cases of good operative risk are not referred to the Radiumhemmet. From 1948 to 1951 they amounted to about 50 per cent of all cases of carcinoma of the corpus diagnosed in two-thirds of the country.

Therapy

The types of treatment employed in carcinoma of the uterine corpus are total hysterectomy by the abdominal or vaginal route, radical hysterectomy, radiotherapy, or a combination of radiotherapy and operation. In the United States and Great Britain there has been an attempt in the last few years to apply radical hysterectomy with bilateral lymphadenectomy to cases of carcinoma of the endometrium because of the reports that metastases to obturator, hypogastric, or iliac lymph nodes should occur in up to 21 per cent of the cases.^{4, 16, 17, 25, 36-38} Our experience does not quite agree with these reports. It is true that metastases to the obturator and hypogastric lymph nodes occur in cases of adenocarcinoma in the corpus deeply infiltrating the myometrium, but I believe it is not a matter of common occurrence. In cases of carcinoma of the corpus and endocervix or isthmus, however, such metastases are rather frequent. We have seen metastases to the pelvic lymph nodes in 4 of 8 patients with carcinoma of the corpus and endocervix but in only one of 50 patients with carcinoma of the corpus operated on after primary irradiation. Metastases to

the aortic glands were also present in the last-mentioned case. As far as operation is concerned, therefore, we feel that radical hysterectomy with lymphadenectomy only may be the treatment of choice in cases of carcinoma of the endometrium with extension to the isthmus or cervix.

A primary hysterectomy by the abdominal or vaginal route gives good results in a patient with a Stage I carcinoma of the corpus considered a good operable risk. We must insist that a presentation of surgical treatment of carcinoma of the corpus be based upon the general rules adopted. It is not justifiable to compare statistics based on surgically classified cases with those based on clinically classified cases, nor is it possible to draw any conclusion from results obtained in a selected series of cases. Recently Furuhjelm¹⁰ has not paid any regard to these facts in presenting the 5 year results obtained by primary surgery in cases of carcinoma of the corpus at one university clinic in Stockholm. The conclusions drawn are misleading since actually only 55 per cent of the Stage I patients have been operated upon and since patients with no histologically proved carcinoma are included in the series.

Recurrences in the vaginal vault or in the vagina are not rare in cases of carcinoma of the corpus primarily operated on, and occur also in patients operated on by the vaginal route and in cases in which suture of the cervix was carried out prior to operation. The incidence of such recurrences is about 12 per cent according to reports from different countries.

At the Radiumhemmet, we have advocated the use of radium as a prophylactic postoperative procedure. Without anesthesia we introduce a cylindric applicator with 100 to 150 mg. of radium element into the vagina and give a dose of 2,500 to 3,000 gamma r to the depth of 1 cm. of the wall. Provided a carcinomatous spot is diagnosed in the vagina, another application of radium is given toward this area 3 weeks later.

Three hundred forty-seven cases of carcinoma of the corpus primarily operated upon and postoperatively treated by irradiation have been the subject of an investigation. The 5 year survival rate is 80.4 per cent, the 10 year rate 69.1 per cent. In 207 of the 347 cases, the carcinomatous infiltration was superficial; the 5 and 10 year survival rate is 90.8 and 80.6 per cent in these cases. The corresponding figures for the deeply infiltrating carcinomas are 63.4 and 53.1 per cent; in our opinion, x-ray therapy also should be applied in those cases. Recurrences to the vaginal vault or to the vagina have occurred later in 15 of the 347 cases (4 per cent).

The same treatment has been applied in 29 cases of early carcinoma of the corpus and endocervix. The 5 year survival rate of 41.4 per cent is not satisfactory.

The value of radiotherapy in adenocarcinoma of the corpus is further demonstrated by the fact that 41 of 102 patients referred to the Radiumhemmet for recurrences appearing after primary surgery have been satisfactorily treated—a 5 year apparent recovery rate of 40.2 per cent.

Patients with carcinoma of the corpus primarily referred to the Radiumhemmet have received irradiation. It is not true that we have stressed that radiotherapy should be the primary treatment of carcinoma of the corpus. We have tried to find a radiation technique which would lead to satisfactory results in patients with endometrial carcinoma who are considered to be poor operative risks or inoperable. For years Heyman¹⁴ had tried various techniques with unsatisfactory results. In 1941 he described the "packing method," i.e., the filling of the uterine cavity with multiple radium sources. In consequence of the improved results Heyman achieved and of the few complications, surgeons and gynecologists in Sweden referred more cases of corpus cancer for primary radiotherapy than previously. Since 1936 the packing method has

been our standard radiation therapy. From 1936 through 1951, 1,439 cases of carcinoma of the corpus, 290 cases of carcinoma corporis et endocervicis, and 131 cases of carcinoma uteri et ovarii have been primarily treated by irradiation. The packing of the uterine cavity has been divided into 2 or 3 applications with an interval of 2 to 3 weeks. Simultaneously with one of the uterine applications, radium is applied to the vagina in accordance with the technique recommended in cases primarily operated upon. If the cancer has extended to the endocervix or if vaginal, usually suburethral, metastases are present, a second application of radium is given toward the portio and/or the area of the metastases. A suitable applicator is used and 1,900 mg. hr. are given. Additional roentgen therapy is administered to the parametrial tissue in these cases.

The principle of the present packing is to fill the uterine cavity satisfactorily with radium tubes and thus decrease the distance from the radium to the neoplasm as much as possible. The first step is to introduce 3 irradiators with a retainer connected to each of them. The irradiators are pushed up against the fundus by means of an applicator resembling a Hegar dilator and are kept in position by the attached retainer. More and more irradiators are then gently pushed up against the fundus. The length of the wires is the same and this makes it possible to check that the irradiators are in the proper position in the fundus. When the fundus of the cavity begins to be filled, the retainers are removed. It is then possible to fill the remaining uterine cavity by using several additional irradiators. A normal uterine wall is relatively nondistensible, but myometrium that has been invaded by a malignant growth may be gently "ballooned out" by careful application of irradiators to the fundus. This is possible if the wires attached to the irradiators already applied are kept taut with the left hand, while another irradiator is insinuated between the ones previously applied. The application should not be done with force. With this technique a perforation is rare and it is possible to thin out the uterine wall in order to achieve a narrow distance between the cancer and the radium applied. No doubt this part of the packing is important. Having thus packed the upper part of the uterine cavity, we pack the remaining cavity similarly. It is a matter of course that irradiators of the same size as those applied in the corpus should be placed in the cervix.

An x-ray is taken to determine the position of the radium. The dosage in the bladder and rectum is measured directly. The irradiators remain in the uterus for varying lengths of time according to the amount of radium applied and the type of irradiator used. The dosage is physically calculated. As a rule we give a total dose of 3,000 gamma r at 15 mm. from the nearest irradiator. Expressing of the dosage in mg. hr. makes no sense.

The radium is removed by the nurse in charge of the ward with the patient in bed. Difficulties in removing the radium very seldom occur. The patient is discharged the next day and is permitted to carry out her usual work.

Results of Treatment

From 1936 through 1951, 1,860 cases of carcinoma of the endometrium have been treated primarily at the Radiumhemmet. Table I gives the total series of these cases. Carcinoma uteri et ovarii have been classified in cases in which it was impossible to tell, clinically or histologically, whether the primary tumor was situated in the endometrium or in the ovary. This diagnosis is justified only provided the ovarian tumor was clinically diagnosed prior to therapy.

Table II gives the 5 year apparent recovery rates in all cases of carcinoma of the corpus and carcinoma of the corpus et endocervix treated by the

technique described, from 1948 through 1951. The requirements for allotting a case to the clinically inoperable or inoperable group have been presented. Two representative cases of these groups will be presented.

Case Classified as Clinically Inoperable (Stage I, 2).—The patient, 67 years old, arrived at the Radiumhemmet in February, 1949. She had observed vaginal hemorrhage for more than one year. The uterus was the size of a fist. The cavity was large and irregular. The patient also had myocarditis, with arrhythmia, a blood pressure of 280 mm. Hg, and diabetes. She was treated by irradiation. At the time of this writing (1957), her general condition is poor, but she is free from cancer.

Case Classified as Inoperable (Stage II).—The patient, 49 years old, arrived at the Radiumhemmet in July, 1949. She complained of vaginal hemorrhage and discharge for one year. Clinical examination showed several metastases in the lower part of the vagina and a uterus the size of a fist fixed to the right pelvic wall by a firm, nodular parametrium. The patient is living and symptom free at the time of this writing (1957).

TABLE I. CARCINOMA CORPORA (1936-1951)

AGE	TOTAL		OPERABLE		INOPERABLE		DIED FROM INTERCURRENT DISEASE
	NO. OF CASES	5 YEAR CURES	NO. OF CASES	5 YEAR CURES	NO. OF CASES	5 YEAR CURES	
<i>Cancer Corporis Uteri.—</i>							
<40	17	14	16	14	1	0	1
40-49	157	135	149	131	8	4	4
50-59	567	413	519	401	48	12	30
60-69	497	275	426	257	71	18	58
70-79	193	71	162	66	31	5	43
>80	8	3	6	3	2	0	1
	1439	911	1278	872	161	39	137
	63.3%		68.2%		24.2%		9.5%
<i>Cancer Corporis et Endocervicis.—</i>							
	290	98	138	64	152	34	5
	33.8%		46.4%		22.4%		-
<i>Cancer Uteri et Ovarii.—</i>							
	131	42					
	32.1%						

TABLE II. CARCINOMA CORPORA UTERI (1948-1951)

	NO. OF CASES TREATED	LIVING AT 5 YEARS WITHOUT EVIDENCE OF CANCER	5 YEAR RELATIVE CURE RATE (%)
Clinically operable	293	248	84.6
Clinically inoperable	177	88	49.7
Inoperable	62	17	27.4
Total	532	353	66.4
Died from intercurrent disease	49 (9.2%)		
Operated on after failure of radiotherapy	77 (14.5%)	51	66.2

Fifty-four of the 617 patients treated from 1948 through 1951 have died from intercurrent disease within 5 years. Most of them belong to the clinically inoperable group (Stage I, 2). In 88 patients, operation was performed within 5 years of presumable failure of radiotherapy. In 21 of these cases, no living cancer could be found in the specimen. If we bear in mind that many of the prognostically favorable cases are primarily operated on and, thus, cannot be included in the Radiumhemmet series, I feel that it is obvious that primary radiotherapy adequately administered is justified as a choice of treatment in carcinoma of the endometrium.

Comment

In 1957 McKelvey³¹ stated that proved tumor outside the uterus has been treated with long focal distance therapy sufficiently often to produce the conviction that no cures are obtained under these circumstances by the maximum dose which one dares to give. He points out that a minimum dosage of 12,000 r is required to sterilize an adenocarcinoma of the endometrium. From our results, it is obvious that we cannot agree with McKelvey. His conclusions are based on results obtained with an unsatisfactory irradiation technique.

It has sometimes been pointed out that the risk of complications with the Heyman packing method should be great. It is true that a perforation of the uterus sometimes is unavoidable. A perforation may be due to the cancer or

Fig. 10.

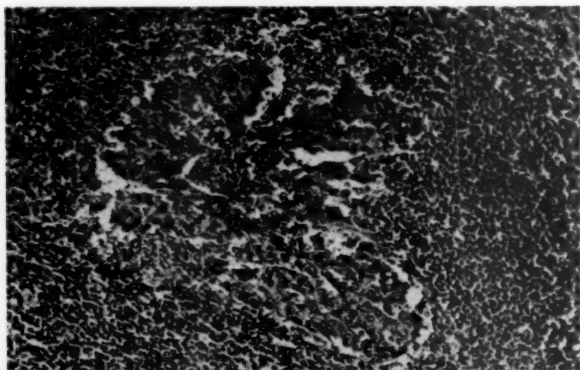


Fig. 11.

Figs. 10 and 11.—Pronounced radiation changes in external iliac lymph node metastases from a patient with advanced carcinoma of the corpus and endocervix. ($\times 160$; reduced $\frac{1}{2}$.)

may occur at the time of curettage or packing. As a rule, it is a harmless complication and does not require any action. A perforation occurred during the packing in 27 of 617 cases treated at the Radiumhemmet from 1948 to 1951. The application of radium was quite easy in most of these cases, but in one instance of advanced carcinoma the irradiator was broken and lost in the peritoneal cavity. A laparotomy was performed and the uterus was removed. The patient died later from cancer. Thirteen out of the 27 patients mentioned are living without any evidence of disease in 1957.

Until recent years surgery at the Radiumhemmet has been performed only in cases of presumed failure of radiotherapy. The reason for this attitude was

the desire to find an irradiation method which would give satisfactory results. Since we now believe the packing method has realized this hope, we have in recent years referred patients of good operative risk to surgery 6 weeks after completion of radiotherapy. This combined method has been recommended by several American authors. Although as yet we cannot present any results, we are convinced that primary radiotherapy according to the packing method followed in 6 weeks by operation will lead to improved results in cases of poorly differentiated adenocarcinoma of the corpus.

In cases of far-advanced carcinoma, neither radical operation nor radiotherapy will give satisfactory results. We have tried various techniques in such instances. We like to start the treatment with the application of a radium tandem into the enlarged cavity. We give 3,000 mg. hr., let the patient turn in bed during the application, do not consider this treatment as far as further dosage is concerned, and start the packing within 2 to 3 weeks.

The following case will give an idea of this type of individualized therapy.

A 64-year-old patient arrived at the Radiumhemmet June 5, 1957. Gynecologic examination showed a large endocervical crater and a fixed uterus, the size of a grapefruit, filled by pyometra and necrotic anaplastic adenocarcinoma. In both iliac fossae a fixed nodular mass, the size of a hen's egg, was palpated. A tandem with 300 mg. of radium was applied immediately into the irregular uterine cavity, which was 12 cm. in length. The radium was left in situ for 12 hours; 2,000 skin r were given to the mass from one anterior portal and, on June 15, 80 mg. of radium element was applied into the vagina for 27 hours. Since the general condition improved considerably and the growth in the pelvis responded satisfactorily, the uterine cavity was packed with multiple radium sources on July 9, and, simultaneously, radium was applied into the vagina. Roentgen therapy was later given from posterior portals toward the pelvis, 2,000 skin r to each field.

In September, 1957, the improvement was obvious, and a radical hysterectomy with lymphadenectomy was performed on September 11. The uterus was the size of a goose egg. The cancer had broken through the posterior wall of the uterus, but there were no peritoneal metastases. The obturator and hypogastric lymph nodes were greatly enlarged and fixed, particularly on the left side. The left internal iliac vessels and external iliac vein had to be removed with the mass. Microscopic examination of the specimen showed an anaplastic adenocarcinoma in the corpus, cervix, and lymph nodes (Figs. 10 and 11). Obvious changes due to the irradiation could be seen in the uterus as well as in the metastases. The patient left the hospital in excellent condition.

Summary

The Radiumhemmet series of carcinoma of the endometrium has been presented in detail. The significance of a careful diagnosis prior to treatment and of a precise classification and staging of the disease has been stressed. The treatment should be individualized with regard to the extension of the disease and the differentiation of the adenocarcinoma. A radical hysterectomy and lymphadenectomy are indicated in cases of carcinoma of the corpus and endocervix or isthmus but are not the treatment of choice in a carcinoma limited to the corpus.

A fractionated curettage or a hystero-graphy with water-soluble contrast medium facilitates a careful diagnosis. The filling of the uterine cavity with multiple radium sources according to the packing method has proved to give satisfactory results in patients with carcinoma of the endometrium who are considered to be poor surgical risks. Surgery and radiotherapy are both imperative in the treatment of carcinoma of the endometrium.

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DEPARTMENT OF CURRENT OPINION

Re-evaluation

NOT in all cases can a Re-evaluation stem from the author of the original remark, the inventor of the first instrument. In some instances we must re-appraise that which comes to us down through the years, and the originator is lost in the shadows of an ill-recorded history.

In the following article Dr. F. H. Howard re-evaluates (and challenges) the recumbent lithotomy position for delivery. He would be the last to insist that his re-evaluation is unbiased. Certainly, however, it is interesting.

Similarly, Dr. Herman Taller re-evaluates (and disputes) the concept that the management of obesity calls for a reduction in calories.

THE PHYSIOLOGIC POSITION FOR DELIVERY

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IT IS probably impossible to approach any re-evaluation of the recumbent position for accouchement unless we completely detach ourselves from our medical training and daily clinical experience. We have behind us a 2-century-old custom, and it would require not only re-evaluation but considerable re-education to achieve a negative attitude toward the recumbent position for delivery.

That some sort of an upright position for the process of childbearing is the position used in antiquity and in the present among primitives is not disputable. Ford,¹ Professor of Anthropology at Yale, in cataloguing the positions used in primitive societies, states: "The woman usually assumes a sitting position; less commonly she either kneels or squats." He goes on: "In our modern hospitals the reclining position has been adopted since it facilitates antiseptic treatment. The evidence from our primitive societies indicates that this is a most unnatural position."

Professor Ford's assertion that antiseptic technique is involved in the retention of the recumbent position will not thoroughly hold water. The inception of the recumbent was a full 100 years before Lister, Pasteur, and their contemporaries, including Semmelweis, brought out the facts of sepsis. That a woman in an upright position is less apt to develop sepsis is almost self-evident. The reason for retention of the recumbent position is entirely the dictate of custom.

Historically, the recumbent position was used by Mauriceau in order to apply the Chamberlen forceps more easily. The Sims position is used on the continent of Europe and to a lesser extent in other places where medical thinking has been influenced by European medical mores.

A normal delivery through an adequate pelvis is in truth a problem in extrusion of a compressible solid, the fetus, through a semirigid aperture, the pelvis, and should be considered as if we were taking a course in undergraduate physics.

By way of comparison, consider the tube of tooth paste. The paste therein is semisolid. When one squeezes the tube, with the long axis of the tube in the horizontal axis, a slow squeeze causes extrusion of the paste. As it emerges into an area of no support, the column of paste bends toward the center of the earth. If one gives this tube a violent squeeze, the column of the paste is apt to be fractured. If, on the other hand, one holds the tube in the vertical position, the same violent squeeze will not fracture the column of paste, or it will not fracture it as badly as in the horizontal position.

When we translate this to the extrusion of a fetus, one might believe it to be logical for the mother, if unattended, to wish to be on all fours, and that is essentially what happens in the case of quadruped animals; the birth canal is angled toward the center of the earth. With humans, however, the assumption of the erect posture gives us an even more logical use of gravity in our birth.

Wylie,² by the aid of a spring mechanism in a Bill axis traction handle, found that the average pull necessary to extract the fetal head is 35 pounds. This is with the mother in a recumbent position. If one assumes an average weight of 7 pounds for the baby, then one may subtract this 7 pounds from the externally exerted force, which makes but 80 per cent of the pull necessary in the recumbent required for delivery in the upright position. A further factor is the efficiency of the expulsive effort. Mengert and Murphy,³ in 1933, reported on the efficiency of the expulsive effort and found that if one considers the Sims position to be 100 per cent efficient, then the sitting position is 130 per cent efficient in expulsion. By adding these factors, one is left with the answer that the coefficient of expulsion is, in the upright, 50 per cent of that necessary in the Sims position. The sitting over the lithotomy position is 54 per cent. This concerns but the forces applied to the fetal head.

The birth canal in the human female is approximately 30 degrees from the vertical, with its inception cephalad, and its caudad reversed approximately 60 degrees. At the end of this reverse curve, extension of the fetal head causes a sharp hook on this final phase in the birth of the head. It is my belief that the actual physiologic position is mobile and varies with different phases of labor and different locations and positions of the fetus. Roughly, the upright position concurs with gravity in all save the extension of the head. A woman in labor might then be walking about for the first stage of labor, be squatting with most of the contractions in the second stage, and then with the extension of the head, while squatting, place her hand so as to catch the baby's

head, and arise. In this last maneuver, she would shift her weight and body forward, leaning further, in order to get up. This last maneuver would keep the plane of expulsion more in line with the plane of the force of gravity. Or, she may fall forward on her knees with one hand as support, and thus again, the plane of the expulsion would be in the plane of gravity.

This sequence of directions would imply a delivery room facility which would shift through a variety of positions. The fact that by the use of a different mode of table for accouchement the operator may have full control of this situation, as well as be comfortable himself, requires no flight of imagination. The "Rube Goldberg" mechanisms in which certain specialists, such as neurosurgeons, position their patients would lead us to believe that a simple mechanism might be employed to position our patients in a physiologic position. As a matter of fact, the table devised by my wife and me is simple; and with a few changes might be simpler and more adequate. The technique is not difficult; it consists of rotating the patient through a 90 degree arc after placing her initially in the lithotomy position. It must be noted that the angle of attack must be near 90 degrees; 45 is not enough for the correct application of force during the extension of the head. The method is applicable to any of the commonly used methods of pain relief save inhalation anesthesia, including hypnosis and the Read method. Where caudal or spinal blocks are used, careful attention must be given to the blood pressure.

In my own experience, all but one of my patients to whom a block anesthesia, recumbent, was given required help in expelling the fetus.⁴⁻⁷ With the upright position, in the neighborhood of 50 per cent were delivered without forceps help. It is my opinion that the upright will improve results of anyone the same way. A recent appraisal of this work in Upjohn's house organ, *Scope*, quotes several of the elite of our specialty as favoring a large-scale trial and evaluation of the physiologic position. This I am in favor of; it should be tried in teaching institutions, not one, but many, and should also be tried in lesser centers.

So it is; I contend that parturition is easier and safer in the upright position. I would not have anyone think that I am adamant in this matter, for we must live with our fellow physicians and our patients. Our present mores as a group are such that conservative thinking in this respect is regarded as being quite radical, and my table has not been allowed in any of the nearby hospitals. Little by little I would hope to convince my colleagues of the worthwhileness of this project. The patients are not hard to convince.

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THE PROBLEM OF OBESITY

HERMAN TALLER, M.D., BROOKLYN, N. Y.

EXCESSIVE weight, rapid weight gain, and initial obesity are frequent problems met in obstetric-gynecologic practice. These call for control and/or reduction of weight in addition to the specific obstetric or gynecologic condition under consideration. Excessive and rapid weight gain in pregnancy predisposes to toxemia and is an added complication in gravid patients with heart disease, diabetes, nephritis, and other related metabolic conditions. In the field of gynecology a close relationship exists between obesity, menstrual irregularities, endocrine disturbances, to a degree, as they relate to infertility, and even postural backaches. This presentation will be limited to a discussion of obesity due to faulty nutritional patterns, dietary irregularity, and poor selection of food elements, the correction of which will have a salutary effect on the obstetric complication and/or on the specific gynecologic problem.

Even at present, it is the general belief that obesity can be controlled by calorie reduction. However, a recent publication by Stunkard and McLaren-Hume¹ refutes this concept completely. In their paper, the authors report that, out of 100 obese patients treated with an intake of between 800 and 1,500 calories daily, after a prolonged period, only 12 per cent lost more than 20 pounds and only one patient lost more than 40 pounds. Maintaining the weight loss of these patients was a difficult problem; and even more difficult was having these patients return to the clinic. Twenty-eight per cent of their patients never returned to the clinic after the initial treatment; 2 years later, only 2 patients had maintained their weight loss. The authors make an important point in calling attention to the ill effects which resulted from this regimen. Among these were emotional disturbances, nervousness, general weakness, and other psychosomatic syndromes. They conclude that some other type of treatment is needed.

Various methods have been used to determine the amount of body fat in a normal individual. Sen and Banerjee² determined the percentage of body fat by using several methods. One of the methods they used was calculation of the specific gravity of the body as determined by water replacement according to the formula of Rathbun and Pace³; another was utilization of the density of the body according to the formula of Keys and Brozek.⁴ They concluded that the average fat content of the body (by means of the specific gravity method) was about 11 per cent of the total body weight; with the second method it was 11.3 per cent. These percentages of body fat were based on figures for ideal height and weight.

To determine the degree of obesity, the tables of the insurance companies for age 25 to 30 are used. These tables were prepared by Dublin and Marks⁵

and indicate the normal weight for persons between the ages of 25 to 30 years. This weight should remain constant throughout a person's life. The amount of weight which is more than 10 per cent above the average weight for a particular height constitutes the degree of obesity. For the woman below the age of 25, one pound should be deducted from the average weight for each year below 25.

In the past few years, much knowledge has been accumulated in the field of nutrition by use of tagged carbohydrates, proteins, and fats. More recently, there has been an increase in the knowledge of the metabolism of fats and the relationship of dietetic fat to adipose tissue, as well as the relationship between carbohydrate intake and the adipose tissue. According to Pennington,⁶⁻⁸ obesity is due to excessive intake of carbohydrates plus a disturbance in its metabolism. He points out that the disturbance lies at the level of the pyruvic acid which fails to oxidize further and as a result accumulates. This accumulation inhibits the oxidation of fats.

Chalmers, Kekwick, and Pawan⁹ have identified a substance which they call "mobilizing factor." This substance has been found in the urine of fasting individuals and also in those people in whom the calorie intake was mainly in the form of fats. A lesser amount was found in the person on a high protein diet, but this substance could not be found in those on a balanced diet or when the calories were supplied mainly in the form of carbohydrates. It was their conclusion that this particular substance, when injected into mice, caused weight loss without depressing the appetite when given over a period of time, the loss being in the form of body fat and water. This is of great significance in the treatment of obesity.

Until recently, before tagged fats had been introduced in research, the concept was that adipose tissue was an inert padding. A research team of the Veterans Administration, headed by Feller and Feist¹⁰ of the Veterans Administration Hospital in Seattle, has found that fat tissue converts carbohydrates into fat at a much faster rate than do other tissues. These researchers have incubated various mouse tissues and have fed these tissues with glucose, acetate, propionate, and methylmalonate, all of which are found in nearly all body cells. All these compounds were "tagged" with radioactive material so that their reactions could be traced biochemically. They concluded that fat was produced more than 100 times as fast by fat as by tissue from the liver. These studies indicate that the chemical pathways by which fat builds fat differ from those used by other tissues. For example, fat makes fat out of acetate, a substance that other tissues convert to a variety of nonfat substances. This would easily explain the vicious cycle of adiposity—the fatter the individual, the faster she will gain weight.

From what has been said, it may be concluded that the treatment of obesity should consist of a change in the eating pattern toward a high fat, high protein, and restricted carbohydrate diet. More recent investigation into fat metabolism has pointed out two great divisions—saturated fats and unsaturated fats. Recent literature has been full of reports concerning the effect of saturated fats upon arteriosclerosis and coronary and general ischemic heart disease, as well as the direct relationship between the dietetic saturated fat and the elevated cholesterol level. Gofman and his co-workers¹¹ went even further by demonstrating not only a correlation between the saturated fats and lipoproteins Sf 0-12 and Sf 12-20, but also the fact that a dietary carbohydrate intake is highly influential in controlling the blood content of those fatty materials known as Sf 20-100 and Sf 100-400 lipoproteins. For this reason, they advocate curtailment of the saturated fat and carbohydrate intake.

The principles of the treatment of obesity should follow those outlined by Pennington,⁶⁻⁸ Kekwick and Pawan,¹² and Thorpe.¹³ More recent reports have

indicated that a diet high in animal fat will elevate the serum cholesterol considerably, sometimes to a dangerous point. Therefore, the animal fat should be replaced by unsaturated fatty acids as described by Ahearns,¹⁴ Sinclair,¹⁵ Hansen,¹⁶ Kinsell and associates,¹⁷ Keys,¹⁸ Brown and Page,¹⁹ and Bronte-Stewart and Blackburn²⁰ to reduce serum cholesterol levels. A newer concept of treating obesity, then, based on a modification of Pennington and Kekwick and Pawan, consists of replacing the dietetic saturated fat as much as possible with unlimited amounts of unsaturated fat, regardless of the increase in calorie intake by the unsaturated fats. A major advantage of this nutrition pattern is its high satiety value.

If the carbohydrate intake is reduced to a minimum, all the pyruvic acid formed will be oxidized. In other words, all the carbohydrates ingested are oxidized into carbon dioxide and water. Restriction of carbohydrates also removes the stimulus of insulin function so that, on one hand, the fat storage activity of the insulin is held to a minimum, and, on the other hand, a low pancreatic secretion will stimulate the anterior pituitary to form more fat mobilizing principle which will predominate the fat storing forces. Mobilization of fat out of the adipose tissue will be greatly enhanced and consequently oxidized to ketones in the liver and circulated to the tissues in an easily combustible form.¹³ No matter what process of fat mobilization is brought into action, ketogenesis must play a vital role, and it is a well-known fact that carbohydrates can be converted into body fat while fat cannot be converted into carbohydrates except in a very small, insignificant amount.

It is very interesting to note Stefansson's²¹ observations among the Eskimos that there was no obesity when their accustomed diet of lean and fat meat was followed,* but obesity appeared with great rapidity when concentrated carbohydrates were introduced into their diet.

The following observations have been made by Kekwick and Pawan: In the plasma of normal fasting humans, 35 per cent of the fatty acids are found as phospholipids, 45 per cent as triglycerides (neutral fats), 15 per cent as esters of cholesterol, and only 5 per cent are nonesterified fatty acids. The nonesterified fatty acids have a very active metabolic role and their utilization is rapid. Their plasma concentration is sharply affected by the administration of insulin or glucose (which causes a fall) and by dextroamphetamines or epinephrine (which causes a rise). The nonesterified fatty acids are composed chiefly of palmitic, stearic, oleic, and linoleic fatty acids. In fasting animals, these substances rise, the source being adipose tissue. On the other hand, intake of carbohydrates inhibits the release of nonesterified fatty acids from fat depots. It is these nonesterified fatty acids that an obese individual has to increase in turnover in metabolism. According to Kekwick and Pawan, a high fat diet will form the mobilizing factor and will stimulate nonesterified fatty acids out of the fat depot. Also, according to Pennington, a reduction of the carbohydrate intake results in a decrease in the concentration of pyruvic acid, thus eliminating its inhibitory action on the formation of the mobilizing factor.

The type of nutrition pattern advocated for the treatment of obesity is that of unlimited amounts of unsaturated fats, high protein, restricted saturated fats, and very restricted carbohydrates. This regimen is continued throughout the period of treatment until the patient reaches her desirable weight according to the insurance company tables. The purpose of this nutrition pattern is to re-establish a balance between the liver and the adipose tissue lipogenesis. When the patient reaches her established weight, 11.3 per cent of her total body weight is represented by adipose tissue (according to Sen and Banerjee). It is at this

stage, when the balance between the liver and adipose tissue lipogenesis is established, that the patient will for the future be free of obesity, provided the carbohydrate intake is metabolized completely. This, of course, can be established later by gradually increasing the amount of carbohydrate intake daily up to the point where weight gain occurs.

I applied these principles in the treatment of markedly obese pregnant women (whose weight when first seen was about 200 pounds) as well as obese women with various gynecologic complaints. A number of these patients were discovered to be diabetics (previously unknown); others had a high blood cholesterol; still others had hyperlipemia or various alterations in clotting and coagulation times. The degree of obesity and the biochemical determinations in regard to the glycemia, cholesteremia, lipemia, protein-bound iodine, and pyruvic acid determinations, together with the results of the blood count, determined the type of nutritional pattern for each patient. The percentages of carbohydrate, protein, and unsaturated fat intake were different in each instance. No effort was made to calculate or restrict calorie intake. Since each patient had to be calibrated individually, a general pattern, as a rule, could not be established because of the fact that the individual tolerances toward oxidation of carbohydrates were so varied and the degrees of obesity among these patients were so different.

After the initial visit and after the biochemical determination reports were returned, a new pattern of eating habits was developed for each particular patient by adherence to the principles described above. (A discussion of the various nutritional patterns will be presented in a subsequent article.) Amphetamines were used, not because of their anorexic effect, but because of their turnover factor of the nonesterified fatty acids from the fat depots, and they were very helpful. Supplements of vitamin B complex and vitamin C were given routinely.

At subsequent visits, at 2 week intervals, blood pressure, weight, and urine were checked routinely. All of the patients' complaints were recorded. Some were purely psychosomatic, but at no time were there any major complaints. Their hunger definitely disappeared, the desire for carbohydrates was still present in a number of patients, and a very small number still craved sweets. At 6 week intervals, a new biochemical determination was done to observe the changes. Wherever there had been hypercholesteremia, the cholesterol level was definitely lower. Also, protein-bound iodine was corrected and a number of anemias, which notably were observed in about 75 per cent of the obese individuals, gradually started to improve.

Summary

1. The theories concerning the etiology and treatment of obesity are re-evaluated.
2. The principle for the treatment of obesity consists of re-establishing the equilibrium between the lipogenesis of the adipose tissue and of the liver. Fundamentally, this requires a diet high in fat and protein but low in carbohydrate. The fats should be vegetable in origin.
3. The treatment consists of first determining the biochemical reactions of the individual patient, and then placing her on an appropriate obesity regime.

Appreciation and thanks are expressed to Dr. William Levine, Director of Obstetrics and Gynecology at Beth-El Hospital, for his advice and guidance in the preparation of this report.

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Erratum

Under Summary and Conclusions, on page 611 of the article, "Sarcoma of the Uterus," by H. Melvin Radman and William Korman, in the September, 1959, issue of the JOURNAL, Paragraph 3 should read: In this 10 year study, 6,349 patients were found to have myomas; 19 had sarcoma of the uterus; 253 had carcinoma of the cervix; and 129 had carcinoma of the endometrium.



Reviews and Abstracts

EDITED BY LOUIS M. HELLMAN, M.D.

REVIEWS OF NEW BOOKS

Lehrbuch der Geburtshilfe. By Prof. Dr. Heinrich Martius, co-author, Prof. Dr. H. Hartl, illustrated by Käthe Droysen. 731 pages, 768 figures. Stuttgart, 1959, Georg Thieme Verlag. \$14.20.

This volume is the fourth edition of the standard textbook of obstetrics in many German-speaking medical schools. The third edition of the work, issued in 2 volumes shortly after the war, was poorly printed though scientifically excellent. The present edition has been brought up to date and its format has been restored to the high quality of the first 2 editions. The volume remains a fine textbook, reflecting the extensive experience of its distinguished author.

Since the last edition, advances in obstetric practice have necessitated fairly extensive revisions of the text together with the addition of several new features. Among these are lucid sections on afibrinogenemia and amniotic fluid embolism, fetal sequelae of maternal diabetes, hypoxemic damage in newborn infants, and neonatal pediatrics. In addition to these new features, the book provides a complete coverage of topics included in the standard textbooks of obstetrics. The book is divided into sections on normal obstetrics, abnormal pregnancy states, coincidental maternal complications, multiple pregnancy, puerperium and lactation, and the physiology and pathology of the newborn.

Of special excellence are the clear descriptions of nidation, fertilization, and basic embryology and endocrinology. American obstetricians may question the volume's emphasis on external mensuration; for example, after giving elaborate directions for measuring Baudeloque's diameter, Martius admits its negligible clinical importance but recommends its measurement routinely as a means of obliging the obstetrician to pay more careful attention to the shape of the pelvis. Surely there must be less wasteful means to this praiseworthy end.

Martius presents excellent tables of the sequential events occurring during pregnancy. The section on medicolegal cases regarding the length of pregnancy reviews the German cases and is an interesting supplement to the English and American cases usually reviewed in textbooks in English. The discussion of extrauterine pregnancy is especially carefully done, and the therapeutic indications are clearly described and illustrated. On the other hand, the discussion of eclamptic toxemia of pregnancy is uncharacteristically superficial and lacking in clinical detail. The section on the mechanism of labor is also incomplete and sketchy. Prof. Martius' exposition of the clinical management of mechanical problems of delivery demonstrates the author's special interest in this subject—an interest he has displayed impressively in his handbook on obstetric operations.

The estimation of pelvic capacity is discussed more from the purely clinical than from the roentgenologic viewpoint; the descriptions of asymmetrically and absolutely

contracted pelves are much fuller than those usually encountered in textbooks written in English, probably because of the tendency of obstetric treatises on this subject in German to draw extensively on the famous Viennese collection of Breus and Kolisko.

There are inevitable controversial points in therapeutic indications, and Prof. Martius' position in such matters does not always coincide with majority opinion in the United States. Other cases in point are his recommendations of the use of Pitocin in placenta previa and his stated belief that the Braxton Hicks version may still have useful applications. A short section on antibiotics in connection with the discussion of the treatment of puerperal sepsis is up to date, and it represents a point of view close to that of current American practice. The section on pediatrics of the newborn is full and well written; especially lucid are the expositions of Rh incompatibilities and the problems of prematurity.

This edition of Prof. Martius' book is beautifully bound in a durable linen cover, and it is clearly printed on excellent paper. The photographs are uniformly good; many of them are new. The drawings of Käthe Droysen, a prominent feature of all previous editions, are retained. In many cases, these are inartistic and even actually misleading. This is especially true of the colored drawings, which often present astonishingly unrealistic representations of the conditions depicted.

Aside from such minor objections as are indicated above, this book represents the matured thinking of one of the world's most distinguished obstetric teachers, and it is a credit to his high standard of pedagogy and his professional eminence. It belongs in the reference library of every obstetrician who reads German. In its new edition, Martius' textbook continues to be an important contribution to obstetric literature.

Longitudinal Studies of Child Personality. By Dr. Alan A. Stone, co-author, Dr. Gloria Cochran Onque. 314 pages. Cambridge, 1959, The Commonwealth Fund by the Harvard University Press. \$5.00.

Drs. Stone and Onque have performed a difficult and onerous chore that will be extremely helpful for all those interested in and studying the development of children. The authors define a longitudinal approach as a "forward-looking approach which aims to study the individual as he passes from one stage to another, and to the influences of succeeding experiences and circumstances." Thus, the authors have reviewed 297 pages that involve longitudinal studies of children. The investigations reported are anterospective and prospective, short term and long term, small series and long series. The bibliography is limited to those investigations concerned with emotional and social behavior in infants and children. Other areas of behavior, such as physiologic, motor, and intellectual, are included only when there is a specified explicit or implicit relation to social and other emotional factors. In each of the studies the complete reference is given and the setting of the study is described. The number of subjects, time span, and methods of observation and testing are also included. Then the findings are summarized and in most instances the original authors' interpretation of the data is given. The papers are essentially limited to those in the English language and the bulk of the papers originate from the Unit. A stated few papers are from France.

This is a valuable work for source material. As in all such works, one may question certain omissions. Thus, papers that are based upon or that are focused upon neurologic studies of the neonate that may reveal brain damage are largely omitted. The sequelae of such injury upon the developing personality and subsequent behavior are well known and significant. Also studies of special groups such as the American Negro do not seem to be adequately represented. In spite of deficiencies, however, this is extremely useful reference work.

Enfermedades de la vulva. By Dr. D. Calandra, co-author, Dr. R. Sammarino. 423 pages, 230 figures. Buenos Aires, 1959, Ediciones M. Segura.

As the authors point out in the prologue, this book was written to fill a gap in the Spanish literature; it is a concise and complete book on diseases of the vulva. The book

is divided into 17 chapters and has the subdivisions of anatomy, benign and malignant tumors, embryology, inflammatory processes, and disturbances in physiology. A special chapter is devoted to the urethral meatus.

The book describes the pathology, etiology, and clinical aspects of diseases of the vulva from original description to modern times. There is nothing startling in the book, but at the same time all available information about each disease entity is clearly stated and easily understood.

The type is clear. The black and white photographs are well reproduced and have been chosen with care by the authors. Unfortunately, the reproduction of the color plates is poor.

Practical Obstetric Problems. By Ian Donald. Second edition, 712 pages, 140 illustrations, 4 tables. Chicago, 1959, Year Book Publishers, Inc. \$11.00.

This second edition of *Practical Obstetric Problems* has undergone extensive alterations and revisions. The author has omitted such subjects as ovulation, menstruation, conception, infertility, diagnosis of pregnancy, and early development of the ovum.

This textbook is to serve as a guide to the practicing clinician. It deals with special cases in obstetrics, such as those of the elderly primipara, the grande multipara, and patients with bad obstetrical histories. The chapter on vomiting, heartburn, and hiatal hernia goes into great detail and points out the importance of these minor disorders.

The new additions of afibrinogenemia, cervical incompetence, vitamin K and its relation with kernicterus in the prematures, x-ray placentography, and pelvimetry are discussed, and the techniques used for conditions requiring details of procedure (forceps, breech extraction, cesarean section, etc.) are discussed.

The chapter on infections in obstetrics is good and it covers the subject adequately. The author's views on puerperal sepsis, breast infections, and thrombosis are of special interest. The outstanding chapter in the text is Antibiotics—Their Use and Misuse. The chapter is broken down into the requirements for successful treatment, the development of resistance to antibiotics, the value and risks of combined antibiotics, and the use of antibiotics in practice. The latter deals with the unit, stability, range of activity antibacterial action, resistance, mode of administration, and toxicity of various antibiotics.

Proceedings of the Second World Congress on Fertility and Sterility. Edited by Dr. G. Tesauero. Volumes I and II, 2,948 pages, 816 illustrations. Naples, Italy, 1956, International Fertility Association.

We have received for review the *Proceedings of the Second World Congress on Fertility and Sterility* which took place in Naples, Italy, on May 18 to 26, 1956. This work was edited by Dr. G. Tesauero, who has done a splendid job. Inasmuch as each volume occupies about 1,500 pages, it is impossible to do an extended review of the proceedings, but the sectional titles are as follows: Endocrine, Metabolic and Hematological Factors in Fertility and Sterility; Endocrine Therapy in Female Sterility; Diagnosis of Ovulation and Its Disorders; Treatment of Disorders of Ovulation; Tubal Physiology and Its Disorders; Evaluation of New Methods of Diagnosis and Medical Treatment of Female Sterility; Occupational, Toxic and Psychological Factors in Fertility and Sterility; Experimental and Clinical Investigations in Female Sterility; Diagnosis of Spermatogenesis and Its Disorders; New Methods of Diagnosis of Male Sterility; Surgery in Male Sterility; and Problems in Animal Reproduction.

The proceedings are presented in 2 volumes totaling 2,948 pages with summaries in the official languages of the Congress: English, French, German, Italian, and Spanish.

Gynecology and Gynecologic Nursing. By Norman Miller and Hazel Avery. Fourth edition, 501 pages, 249 drawings. Philadelphia, 1959, W. B. Saunders Company. \$5.00.

Nurses desirous of adding quality to the care they minister to female patients are advised to read this volume. The all-inclusive content is arranged in a logical fashion,

with each entity receiving its due emphasis. Considered in the content are those common, everyday problems with which the public expects nurses to be acquainted. Stress is placed on the application of fundamental nursing principles to the gynecological patient. Those basic nursing procedures essential to gynecology are reviewed in detail and supported by clear illustrations. The "total patient" is interwoven throughout most of the content. All aspects composing total care are discussed in a thought-provoking manner. This text merits a place on the reserve shelf in the nursing library.

Diseases of Women. By Dr. F. W. Roques, co-authors, Dr. John Beattie and Dr. Jos. Wrigley. Tenth edition, 556 pages, 209 figures. Baltimore, 1959, Williams & Wilkins Company. \$8.00.

A group of 10 leading British obstetrician-gynecologists have collaborated to produce the tenth edition in 40 years of this concise, yet thorough, textbook of gynecology. Its popularity over these years is probably due to its easy readability and its thoroughness.

The book is well organized and each subject is systematically covered in regard to definitions, pathology, symptoms, diagnosis, and treatment. The first portion of the book extensively reviews the anatomy and physiology of the female reproductive tract but at times becomes somewhat repetitious. The gamut of gynecological disorders is covered, and includes a well-written summary of the menstrual disorders and a chapter on urological disorders in the female. The book concludes with a short presentation of the gynecological operations with descriptions of the techniques sufficient for the student to orient himself to the procedures. Photographs are noticeable by their absence and the illustrations are a bit scanty, being limited to schematic drawings.

The one major failing is the frequent use of categorical, matter-of-fact statements, with no documentation. As a result, the reader is left with the feeling that everything in the book is just so, and nowhere is the possibility of controversy or question brought out. This is further underscored by failure to include anywhere in the book a single reference to any literature or experimental work and the failure to note which of the authors is responsible for which section.

A Handbook of Obstetrics and Gynecology for Nurses. By Dr. D. Clyne. 204 pages, 53 figures. Baltimore, 1958, Williams & Wilkins Company. \$4.00.

The objective of compiling a "tidily tabulated answer book" has been fulfilled by the author. Outstanding is the use of numerous, simple sketches which explain many facts presented, instead of having them buried in a jungle of obscure verbiage. The content of this handbook emphasizes the technical and physical aspects, that is, the condition presented by the patient rather than the patient with a condition. Present-day nursing philosophy supports the latter viewpoint. This book is helpful to those nurses seeking a quick reference concerning such factual data as definition of terms, causes of conditions, clinical manifestations, and methods of treatment.

Reproductive Physiology. By A. V. Nalbandov. 271 pages, 38 figures, 56 tables. San Francisco, 1958, W. H. Freeman and Co. \$6.75.

A compilation of lectures on the physiology of reproduction of animals prepared for students of agriculture, zoology, and veterinary and human medicine are presented. There are 12 chapters dealing with the comprehensive problems of sex, including the anatomy and physiology, with emphasis on the endocrine system in relation to these subjects.

The photomicrographs, gross photographs, schematic drawings, and graphs are exceptional, and, for a text, unusually clear and easily comprehensible.

To the practitioners concerned with human fertility and sterility, this informative book is a strong incentive to those who do not always see the effects of their therapy and who might question the potency of their armamentarium. The bibliography is accurate and simplified. To all, even those with only curiosity in the subject, this book is strongly recommended.

BOOKS RECEIVED FOR REVIEW

- Analgesie psychologique en obstetrique.** By P. Aboulker, L. Chertok, and M. Sapis. 172 pages, 17 tables, 3 figures. Paris, 1959, Pergamon Press. 3.500 Fr.
- Behandlung Entzündlicher Genitalerkrankungen der Frau.** By Gustav Döderlein. 148 pages, 23 figures, 2 plates. Leipzig, 1959, Georg Thieme Verlag. DM 15.20.
- Biological Psychiatry.** By Jules H. Masserman. 338 pages. New York, 1959, Grune & Stratton, Inc. \$9.75.
- Cancer. Part IX—Radiotherapy.** By Ronald W. Raven. Volume 5, 425 pages, 136 figures, 45 tables. London, 1959, Butterworth & Co., Ltd. \$18.00.
- Diseases of the Nose, Throat and Ear.** By Chevalier Jackson and Chevalier L. Jackson. Second edition, 886 pages, 1,193 illustrations. Philadelphia, 1959, W. B. Saunders Company. \$20.00.
- Evaluation of the Pelvis in Obstetrics.** By Charles M. Steer. Second edition, 131 pages, 57 figures, 9 tables. Philadelphia, 1959, W. B. Saunders Company. \$4.00.
- The Functions of the Endocrine Glands.** By Peter F. Hall. 290 pages, 77 figures, 8 tables. Philadelphia, 1959, W. B. Saunders Company. \$5.75.
- The Great Pulse.** By Mary W. Standlee. 192 pages, 58 illustrations. Tokyo, 1959, Charles E. Tuttle Co. \$4.50.
- Hernia.** By Sir Heneage Ogilvie. 135 pages, 51 illustrations. Baltimore, 1959, Williams & Wilkins Company. \$6.50.
- Hormontherapie in der Frauenheilkunde—Grundlagen und Praxis.** By Joachim Ufer. 157 pages, 66 figures. Berlin, 1959, Walter de Gruyter & Co. DM 32.
- A Manual of Anaesthetic Techniques.** By William J. Pryor. Second edition, 228 pages, 75 figures, 1 table. Baltimore, 1959, Williams & Wilkins Company. \$7.00.
- Maternal and Newborn Care.** By Frederick H. Falls. 64 pages, 23 figures. Chicago, 1958, E. I. Donahue Publishing Co. \$1.25.
- Il miofibroma dell'utero—dal punto di vista patogenetico.** By Espedito Moracci. 223 pages, 31 figures. Pavia, 1959, A. & L. Garzanti Libreria Internazionale.
- Office Gynecology.** By J. P. Greenhill. Seventh edition, 572 pages, 145 figures. Chicago, 1959, Year Book Publishers, Inc. \$9.00.
- Portio-Karzinom—Frühdiagnose—Morphologie—Genese.** By Kurt Michalzik. 211 pages, 269 figures. München and Berlin, 1959, Urban & Schwarzenberg. DM 36.
- Psychosomatic Methods in Painless Childbirth.** By L. Chertok, translated by D. Leigh. Second edition, 260 pages. New York, 1959, Pergamon Press. \$6.50.
- Radiation, Biology and Cancer.** Edited by Staff of University of Texas, M. D. Anderson Hospital and Tumor Institute. 493 pages. Austin, 1959, University of Texas Press. \$8.50.
- Radiation Therapy.** By Walter T. Murphy. 1,041 pages. Philadelphia, 1959, W. B. Saunders Company. \$25.00.

SELECTED ABSTRACTS

Acta Geneticae Medicae et Gemellologiae

Vol. 8, January, 1959.

Koch, Gerhard: Genetics of Microcephaly in Man, p. 75.

Perosa, L., Ramunni, M., Manganelli, G., and Bini, L.: Considerations on a Case of Homozygous Twins Affected by Sickle-Cell Anemia, p. 87.

A. M. A. Archives of Surgery

Vol. 78, January, 1959.

*De Santo, D. A., Bullock, W. K., and Moore, F. J.: Ovarian Cystomas, p. 98.

De Santo, Bullock, and Moore: Ovarian Cystomas, p. 98.

Using pathological specimens from the Los Angeles County Hospital and Mercy Hospital in San Diego, Calif., the serous and "mucinous" cystomas were investigated and compared statistically after correcting for age distribution in the two groups. Only material from white patients was used. In questionable cases new sections were cut and various additional staining techniques were used.

There were 957 ovarian neoplasms, of which 591 were serous and "mucinous" cysts; this constituted 60.7 per cent of the group. There were 189, or 32 per cent, malignant tumors in this group; 402, or 68 per cent, were benign. A comparison of these neoplasms in Negroid and Caucasian groups with the usual statistical corrections showed no significant differences.

Certain conclusions were drawn from the charts illustrating the incidence of bilaterality, malignancy, and age distribution. They were: (1) in Caucasian women cystomas tend to be more malignant with increasing age whether the tumor is unilateral or bilateral; (2) it tends to be more frequently malignant when bilateral in each of the age groups; (3) with increasing age there is an increased tendency toward bilaterality; (4) contrary to most writers who state that serous cystomas are more frequently bilateral, bilaterality is not significantly different in serous and in mucinous tumors of comparable age; (5) contrary to the general information, it was shown that the incidence of malignancy in unilateral serous and "mucinous" cystomas was the same except in the 50- to 69-year-old group, which the writers attributed to "sampling error"; (6) in bilateral tumors of the cystomas there were no differences in the incidence of malignancy in the two groups.

In the second part of this communication, 9 cases of the rare ovarian adenoacanthoma were added to the literature. Eight of these were traceable to papillary serous neoplasms and one to an endometrial cyst. Although in 6 cases intrauterine adenoacanthoma was also present, the authors believe that they show evidence that the lesion was primary to the ovary. The mesonephroma of Schiller and the hypernephroid carcinoma with clear cells of Saphir and Lackner are likewise traceable to serous adenoacanthoma and represent variants, according to those authors.

A. STARK WOLKOFF

*This article has been abstracted.

February, 1959.

*Hurwitt, E. S., and Lebendiger, A.: Ectopia Cordis in a Twin, p. 197.

Hurwitt and Lebendiger: Ectopia Cordis in a Twin, p. 197.

This is a case report of a 34-year-old white nullipara who gave birth to apparently monozygotic twin boys weighing approximately 1,900 grams each. The younger twin was and is apparently normal. The older twin was born with multiple defects, the protrusion of the heart apex at a perpendicular angle from the chest being the most remarkable. A surgical attempt was made to close this defect of the anterior chest wall, which extended from the base of the neck to the umbilicus, by mobilizing chest skin. The infant died 12 hours after birth. The congenital defect included bilateral superior venae cavae, interauricular septal defect, interventricular septal defect, an atresic common pulmonary vein, and, of course, a patent ductus arteriosus and hypoplastic left ventricle.

A brief review of the literature is given, and a classification of ectopia cordis is reiterated. A point is made refuting the dictum of MacFarland and Meade that no such defect in a homologous twin occurs without a similar lesion in the other. According to Kindred the blighting probably represents an instance of purely environmental factors acting selectively on one embryo.

The photographs of the heart lesion and the operation plus the schematic breakdown of the anomaly are commendable.

A. STARK WOLKOFF

Boletin de la Sociedad de obstetricia y ginecologia de Buenos Aires

Vol. 37, November, 1958.

*Garcia, C. G., and Constantino, N.: Cancer of the Endometrium in Young Women, p. 305.

Garcia and Constantino: Cancer of the Endometrium in Young Women, p. 305.

The authors present 2 cases of carcinoma of the endometrium in young women. Both the patients were being investigated for infertility. The first patient was 32 years old and the diagnosis was made at the time of hysterectomy. The second patient was 25 years old and the diagnosis was made at the time of endometrial biopsy. Both patients were thought to have had anovulatory bleeding secondary to hormonal dysfunction. The authors also present a cursory review of the literature on endometrial carcinoma in women under 40 years of age.

FRANCIS B. O'BRIEN, JR.

Canadian Medical Association Journal

Vol. 79, Nov. 15, 1958.

*Schneiderman, C., and Stream, G. I.: Vesicovaginal Fistula, p. 801.

Schneiderman and Stream: Vesicovaginal Fistula, p. 801.

Six cases of vesicovaginal fistula are presented with complete healing of the fistula on the first operative attempt. None had previously been operatively repaired and all developed following total abdominal hysterectomy.

The technique advocated is essentially a suprapubic approach to the fistula with extraperitoneal dissection and mobilization. The bladder wall is then bisected well down to and surrounding the fistulous opening and the latter is completely excised. The bladder wall is then mobilized with meticulous care and separated from the vagina sufficiently to allow closure. The edges of the vaginal orifice are closed with interrupted sutures of No. 2-0 chromic catgut everting the mucosa into the vagina. The bladder wall is then closed from below upward with interrupted sutures of No. 2-0 chromic through the muscular wall only. A second reinforcing layer of sutures is then inserted as well as a layer of interrupted No. 3-0 plain catgut sutures through the mucosal layer in the lowermost portion of the bladder.

This technique allows for excellent exposure of the fistula with adequate mobilization of the walls surrounding the fistula without undue tension. This avenue of approach is applicable to the fistulous tract situated above the interuterine ridge and opening high in the vaginal fornix. This is the usual site following total abdominal hysterectomy. The vaginal route appears more suited to the treatment of urethrovaginal fistula or one in which the vesical neck is involved, as occurs after obstetrical trauma.

A complete urological study by intravenous urography and cystoscopy should be carried out to determine the exact site of the fistula. There should be an elimination of infection by cultures of the urine and administration of appropriate antibiotics. A waiting period of 2 or 3 months should precede attempted repair of the fistula in order to allow for regression of edema and infection of the fistulous orifice. There should be continuous and effective postoperative drainage for 12 to 14 days by suprapubic tube without a Foley catheter to allow solid healing of the bladder. Postoperative prophylactic administration of antibiotics should be instituted.

JOHN J. DETTLING

Gazette of the Egyptian Society of Gynaecology and Obstetrics

Vol. 9, January, 1959.

Zayat, M. A., and Gaafar, A.: Dysgerminoma Ovarii (Report of 3 Cases), p. 35.

Wahby, Osman: Trial Forceps and Its Place in Modern Obstetrics, p. 59.

Seif-El-Dine, Diaa: The Results of a Modified Technique for the Treatment of Vesico-Vaginal Fistula, p. 63.

Journal of the American Medical Association

Volume 169, Feb. 7, 1959.

*Harris, J. M., and Nessim, J. A.: To Do or not To Do Cesarean Section, p. 570.

Harris and Nessim: To Do or not To Do a Cesarean Section, p. 570.

During a period of 21 years at the Cedars of Lebanon Hospital in Los Angeles, there were 40,423 deliveries performed, and there were 4,397 cesarean sections. The maternal mortality of cesarean sections was 0.081 per cent as compared with 0.052 per cent for vaginal deliveries. In 1,826 elective repeat cesarean sections, there was no maternal deaths and a perinatal mortality of 1.7 per cent.

The authors stress the importance of an adequate nursing service, blood antibiotics, qualified anesthetists, and pediatricians. Regional block has been the anesthetic of choice. The low cervical cesarean sections were most commonly done by means of a modified Pfannenstiel incision.

Indication for cesarean section, listed in the order of frequency, is as follows: previous cesarean section, fetopelvic disproportion, uterine bleeding, fetal distress, diabetes, uterine inertia, pelvic pathology, toxemia, and previous uterine myomectomy.

Cesarean section provides a safe and easy substitute for those manipulative procedures which have so often resulted in damage to the mother and child. The decision to do or not to do a cesarean section should be influenced only by the welfare of the mother and baby.

JOHN J. DETTLING

Feb. 21, 1959.

Adelman, M., Bell, D. W. J., Giunta, F., Barrett, J. T., Bellin, L. B., and Appleton, R.: Practical Management of Erythroblastosis Fetalis in a Community Hospital, p. 825.

March 7, 1959.

*Flowers, C. E., Jr., Weinell, W. H., and Kirkland, J. A.: Perinatal Mortality in the North Carolina Memorial Hospital, 1952-1958, p. 1037.

Flowers, Weinell, and Kirkland: Perinatal Mortality in the North Carolina Memorial Hospital, 1952-1956, p. 1037.

A study of the perinatal mortality on 2,728 deliveries conducted in the North Carolina Memorial Hospital showed the principal causes of death to be hemorrhagic complications, toxemia of pregnancy, congenital malformations, and premature rupture of the membranes. During the period of study, the perinatal mortality was reduced from 38.67 to 23.39 deaths per 1,000 births for infants weighing 1,000 grams or more.

The highest perinatal mortality occurred among the lower socioeconomic group of patients, whether the classification was based on the mother's education, on race, or on the father's occupation.

Malformations accounted for the greatest number of infant deaths among the private patients while toxemia of pregnancy and premature rupture of membranes accounted for the greatest number of infant deaths among the nonregistered patients.

The incidence of cesarean section for the 5-year period for a total of 2,700 deliveries, was 58, or 2.15 per cent. The incidence among the staff patients (1,717) was 27, or 1.57 per cent, and for the private patients (983) was 31, or 3.15 per cent. The authors stress that the incidence among staff and private patients should be about equal.

The principal errors in obstetrical practice were related to the management of fetal distress, toxemia of pregnancy, and diabetes. The major problems of the neonatal period, as judged from autopsy findings or clinical diagnosis in 63 cases of neonatal death, were prematurity (26 cases), birth injury (13 cases), and infection (9 cases). A reduction in perinatal mortality can occur with improved obstetrical care and with an improvement in the socioeconomic status of the obstetrical patients.

JOHN J. DETTLING

Obstetrics and Gynecology

Vol. 13, January, 1959.

***Biskind, J. I., and Herman, I.: Vitamin K: Its Relationship to Unexplained Jaundice in the Newborn, p. 41.**

Biskind and Herman: Vitamin K: Its Relationship to Unexplained Jaundice in the Newborn, p. 41.

The authors quote several investigators' evidence of the production of hemolytic anemia, hemoglobinuria, and hyperbilirubinemia by the intramuscular administration of large doses of vitamin K. A comparative study was made of infants who developed jaundice early in life born of mothers who received vitamin K and of others born of mothers who did not receive vitamin K.

It was shown in the group of infants born of mothers who received intramuscular vitamin K that there is a direct relationship between the time interval and the level of the bilirubin. Levels were highest for infants born of mothers who received vitamin K less than 10 hours prior to delivery. Levels in premature infants were significantly higher.

The levels of bilirubin were significantly lower in those infants whose mothers did not receive vitamin K at all.

The authors state that, since doses of 1 or 2 mg. of vitamin K given intramuscularly at birth to infants will safely protect them from hypoprothrombinemia, the practice of routine administration of vitamin K to mothers should be discontinued as there is possible danger to the infant whenever delivery is within a few hours after admission of vitamin K. This is especially true for premature infants.

If, on the other hand, the vitamin K is given to the mother to protect her from hypoprothrombinemia doses should be approximately 5 mg. This dose is to be withheld if delivery is imminent.

LAWRENCE J. SONDERS

Zentralblatt für Gynäkologie*Vol. 80, Nov. 1, 1958.*

- *Knopp, K.: The Treatment of Primary Carcinoma of the Urethra in the Department of Gynecology at the University of Würzburg in the Last 30 Years, p. 1705.
Michalowski, E.: On Complete Urethral Plastic Operation, p. 1714.
*Wertsch, H.: The Treatment of Postoperative and Postpartum Urinary Retention With Dihydroergotamine, p. 1718.
Schwarz, P., and Biedermann, G.: Polycystic Kidney and Pregnancy—Case Report, p. 1723.
*Wenig, H.: Follow-up of Vesicovaginal Fistulae, p. 1728.
Wenig, H.: Follow-up of Ureterovaginal Fistulae, p. 1735.
Arabin, H.: Urethral Pessary for the Treatment of Urinary Incontinence, p. 1749.

Knopp: Treatment of Primary Carcinoma of Urethra in Department of Gynecology at University of Würzburg in Last 30 Years, p. 1705.

There were 9 cases of carcinoma of the urethra at the University of Würzburg in the past 30 years. This represents 0.19 per cent of malignancy patients in the Department of Gynecology. Five of the 9 had a 5 year cure. The incidence of this disease is probably higher than generally reported, since in the past most urethral carcinomas were lumped together with vulvar lesions. The present therapy for carcinoma of the urethra consists of radium needles, for about 7,000 r to the center of the tumor followed by x-ray locally, by vaginal cone—5,000 r in 25 to 30 days, and to the inguinal lymph glands for 1,500 r.

WALTER F. TAUBER

Wertsch: Treatment of Postoperative and Postpartum Urinary Retention With Dihydroergotamine, p. 1718.

Fifty patients with postpartum or postoperative urinary retention were treated with dihydroergotamine. Patients who were unable to void spontaneously on the first day after operation or delivery were given 1 mg. of the medication intramuscularly. If necessary, this was followed by 2 mg. twice a day orally. Forty-two of the patients (84 per cent) voided well within 1 day of the onset of treatment. Six of the failures were due to operative trauma. The 2 other patients had severe emotional problems. Urinary retention lasted 3 to 4 days in the control series.

WALTER F. TAUBER

Wenig: Follow-up of Vesicovaginal Fistulae, p. 1728.

Sixty-seven patients with vesicovaginal fistulas were seen in the Department of Gynecology of the University of Greifswald between Jan. 1, 1946, and Sept. 30, 1957. Forty-five fistulas occurred following operations in the department. Fifteen patients had no previous surgery and 7 patients were originally seen for fistulas. Forty-one patients were cured by single operation. Two further patients were cured by a second operation. There were 2 surgical deaths. There were 22 failures, including 18 "gynecological with carcinoma," 2 "gynecological without carcinoma," and one obstetrical.

WALTER F. TAUBER

Nov. 8, 1958.

- *Hollenbach, C.: Clinical Study of the Efficacy of Synthetic Oxytocin, p. 1760.

Hollenbach: Clinical Study of the Efficacy of Synthetic Oxytocin, p. 1760.

Synthetic oxytocin (Syntocinon-Sandoz) was used in 380 patients, for induction of labor, for correction of dyskinesia, during "crowning," for postpartum hemorrhage, for

retained placenta, for uterine atony, for enhancement of lactation, and for treatment of abortion. The synthetic product was found equal to purified posterior extract in all cases.

WALTER F. TAUBER

Nov. 15, 1958.

*Mey, R.: Hyperemesis Gravidarum as Possible Exogenous Cause of Congenital Anomalies, p. 1785.

John, H.: Our Experience With the New Steroid Anesthesia, p. 1791.

Reiffenstuhl, G.: Primary Results of Vaginal Hysterectomy in the Department of Gynecology of the University of Graz Between 1948 and 1954, p. 1800.

Szendi, B.: Giant Sarcoma of the Breast—Case Report, p. 1808.

Mey: Hyperemesis Gravidarum as Possible Exogenous Cause of Congenital Anomalies, p. 1785.

Ninety-eight women with intractable vomiting of pregnancy had 6 babies with congenital anomalies (6.1 per cent). These included babies with congenital heart disease, one of whom died on the thirty-seventh day of life. There were 3 stillborn infants including one with anencephalia, one with harelip, and one with spina bifida. The malformations were thought to be due to glycogen depletion of the mother which leads to disturbance of the carbohydrate metabolism in the embryo during differentiation of tissues. It is, therefore, advised that hyperemesis be treated early and vigorously, before malformations, due to disturbed metabolism, can occur.

WALTER F. TAUBER

Nov. 22, 1958.

Helbing, W.: Twenty-two Years of Treatment of Carcinoma of the Cervix at the University of Jena, p. 1817.

Foukas, M.: Is It Proper to Speak of Toxemia in Premature Separation of the Placenta?—Case Report, p. 1826.

Zinser, H. K., and Bachmann, F. F.: Steroid Anesthesia in Obstetrics and Gynecology, p. 1831.

Krais, W., and Dippon, M.: Spinal Anesthesia Without Headaches, p. 1841.

Gierdal, M., and Butters, G.: Experience With Anesthesia in Old Women Undergoing Gynecological Surgery, p. 1846.

*Cekon, F., and Ehrlich, H.: Parallel Investigation of Pregnanediol and 17-Hydroxycorticosteroid Excretion During Labor and Postpartum, p. 1851.

Cekon and Ehrlich: Parallel Investigation of Pregnanediol and 17-Hydroxycorticosteroid Excretion During Labor and Postpartum, p. 1851.

Pregnanediol excretion was studied in 25 patients. The values were relatively low on the day of delivery and dropped further in the puerperium. No rise was found immediately ante partum. The highest titers occur between the two hundred and fiftieth and two hundred and sixtieth day of pregnancy. The drop thereafter is probably due to aging of the placenta. The 17-ketosteroid excretion was found elevated by 30 to 77.5 per cent in 11 of 16 patients on the day of delivery. Postpartum rise (alarm reaction) was found only following 2 operative deliveries. The baby's sex had no influence on 17-ketosteroid titers. It is suggested that initiation of labor is mediated by the steroid hormones through their effect on intracellular and extracellular concentration of sodium and potassium, which in turn affects muscle irritability and contraction.

WALTER F. TAUBER



Items

American Board of Obstetrics and Gynecology

The Part I Examinations of the American Board of Obstetrics and Gynecology are to be held in various parts of the United States and Canada on Friday, Jan. 16, 1960, at 2:00 P.M.

Candidates notified of their eligibility to participate in Part I must submit their case abstracts within 30 days of notification of eligibility. No candidate may take the Written Examination unless the case abstracts have been received in the office of the Secretary.

Current Bulletins outlining present requirements may be obtained by writing to the Secretary's office.

ROBERT L. FAULKNER, M.D.
AMERICAN BOARD OF OBSTETRICS
AND GYNECOLOGY
2105 ADELBERT ROAD
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International Society of Geographical Pathology

The chief topic of the Seventh Conference, to be held in London, June 29—July 1, 1960, is "Eclampsia and Pre-eclampsia." Members of the Society or other persons wishing to have papers placed on the program should send the title of their paper and a 250 word abstract to the General Secretary, in time to reach him by Dec. 30, 1959. Papers concerned with the chief topic of the Conference or papers of general geographical-pathological interest are suitable.

PROFESSOR FRED C. ROULET
GENERAL SECRETARY
INTERNATIONAL SOCIETY OF GEO-
GRAPHICAL PATHOLOGY
SCHÄUBLIN-STRASSE 17
BASLE 24, SWITZERLAND

Venezuelan Congress on Obstetrics and Gynecology

The Second Venezuelan Congress on Obstetrics and Gynecology organized to celebrate the twentieth anniversary of the Obstetrical and Gynecological Society of Venezuela will be held in Caracas, Feb. 19 to 24, 1960.

For further information write to the Secretary.

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